ENDOCRINOLOGY

The Bulletin of
The Association for the Study
of Internal Secretions

PUBLICATION BOARD

R. G. HOSKINS, *Editor-in-Chief*

MILTON O. LEE — OSCAR RIDDLE

HANS LISSER — DAVID P. BARR

FRANCIS M. POTTENGER

COLLABORATORS

FULLER ALBRIGHT
I. ANDRUSSIER
WAYNE J. ATWELL
GEOFFREY BOURNE
ISRAEL BRAM
JOHN C. BURCH
JOHN C. DONALDSON
MAURICE H. FRIEDMAN
MURRAY B. GORDON
ARTHUR GROLLMAN
FRANK A. HARTMAN
E. C. HAMBLEN
F. C. KOCH
EDWARD LARSON
J. T. LEWIS
JOSEPH M. LOONEY
WARREN O. NELSON
J. P. PRATT
A. T. RASMUSSEN
ELMER L. SEVRINGHAUS
L. B. SHPINER
D. L. THOMSON
R. L. ZWEMER

VOLUME 20

1936

As the Births of Living Creatures, at first, are ill shapen: so are all Innovations, which are the Births of Time.—Francis Bacon, *Essaye of Innovations*

Copyright, 1936, by The Association for the Study of Internal Secretions, Inc. All rights reserved

Composed, Printed and Bound by
The Collegiate Press
George Banta Publishing Company
Menasha, Wisconsin

medical lib
Wahr

CONTENTS OF VOLUME 20

NO. 1, JANUARY, 1936

Action of Anterior Pituitary Hormones on Basal Metabolism of Normal and Hypophysectomized Pigeons and on a Paradoxical Influence of Temperature. *Oscar Riddle, Guinevere C. Smith, Robert W. Bates, Clarence S. Moran, and Ernest L. Lahr* 1
Uterine Response to Pitocin. *Milo R. White and J. P. Pratt* 17
Studies on Ovarian Dysfunction. III. The Menopause. *Fuller Albright* 24
Experimental Production of Ovarian Refractoriness to Anterior Hypophyseal Stimulation in the Monkey. *F. L. Hisaw, Roy Hertz, and H. L. Fevold* 40
The Effects of the Two Ovarian Hormones on Castrated Human Females. *C. A. Elden* .. 47
The Calorigenic Action of Extracts of the Anterior Lobe of the Pituitary in Man. *W. O. Thompson, S. G. Taylor, III, P. K. Thompson, S. B. Nadler, and L. F. N. Dickie* .. 55
Endocrine Dwarfism. *Robert L. Schaefer* 64
The Clinical Significance of Electrical Impedance Determination in Thyroid Disorders. *J. W. Horton, A. C. VanRavenswaay, Saul Hertz, and G. W. Thorn* 72
Fundamental Similarity in the Development of Gonadotropic Response in the Immature Guinea Pig and Rat. *S. C. Freed and A. Coppock* 81
Chronic Hypoglycemia. *Max A. Goldzieher* 86
The Influence of Cortico-Adrenal Extract on the Course of Bacterial Intoxications in Guinea Pigs. *Richard W. Whitehead and Charles A. Fox* 93
Reid Hunt Reaction and the Thyrotropic Hormone. *Hans Wiesbader* 100
An Investigation of the Hormone Content of Saliva. *Abner I. Weisman and Charles C. Yerbury* 103
Clinical and Laboratory Notes
Obesity and Ocular Symptoms in Mentally Alert Children Due to Hypothyroidism. *Jos. P. Costello* 105
Vitamin E and the Gonads. *William Saphir* 107
Short Stature of Patient—A Cause of Error in Performing Plummer's Test for Quadriceps Weakness in Exophthalmic Goitre. *Israel H. Marcus* 109
Current Endocrine Literature 110

NO. 2, MARCH, 1936

Metabolic Studies of Pituitary Insufficiency. *Harold A. Bulger and David P. Barr* 137
Cachexia Hypophyseopriva (Simmond's Disease) with Thyroid and Suprarenal Insufficiency. *Edward Rose and George Weinstein* 149
Pituitary Cachexia (Simmond's Disease). *Robert C. Moehlig* 155
The Effect of an Alkaline Extract of the Anterior Hypophysis upon the Weight of the Spleen and Adrenal Glands and upon the Blood Calcium Level. *Harry B. Friedgood* 159
The Influence of Endocrine Gland Preparations on Allergic Reactions. (Anterior Pituitary Extract, Pregnancy Urine Extract, Follicular Hormone). *Bruno Solomonica and Raphael Kurzrok* 171
Familial Hemolytic Icterus Associated with Endocrine Dysfunction. *Ernest H. Falconer* .. 174
Excretion of Prolan in Essential Hypertension. *Maxwell Scarf and S. Leon Israel* 180
Chemical Changes in the Muscle of the Hypophysectomized Toad. *A. D. Marenzi* 184
Is the Pressor Effect of Glycerin Extract of Adrenal Glands Due to Epinephrine? *R. G. Hoskins and J. S. Gottlieb* 188
The Difference of Response of Males with Undescended Testes to the Water Soluble (Anterior Pituitary-Like) Fraction of Pregnancy Urine. *H. S. Rubinstein* 192
The Effect of Gonadectomy on the Thyroid Gland in the Guinea Pig. *A. A. Kippen and Leo Loeb* 201
The Induction of an Acid Vaginal Secretion in the Immature Macaque by Injections of Oestrin. *B. Vincent Hall and Robert M. Lewis* 210
The Reaction of Fish to Sex Hormones. *Seward E. Owen* 214
The Effect of Injections of Antuitrin-S on the Sexually Inactive Male Ground Squirrel. *Burton Lowell Baker and George Edwin Johnson* 219
Clinical and Laboratory Notes
A Simple Method for the Extraction of Both Estrin and Gonadotropic Hormones from a Single Blood Sample. *S. C. Freed* 224
Studies of Paget's Disease II. The Effect of Adreno-Cortical Extract Treatment upon the Blood Phosphatase in Paget's Disease. *Louis Berman* 226

Squamous Metaplasia in the Cervical Glands of the Monkey Following Oestrin Administration. *Frederick L. Hisaw and Frederick C. Lendrum* 228
Editorial
Practical Sex Hormone Therapy 230
Species Differences in Hormone Production 232
Twentieth Annual Meeting 233
Current Endocrine Literature 235

NO. 3, MAY, 1936

Progestin Content of Blood. *Paul W. Bloch* 307
Studies of the Nature of Antigonadotropic Substances. *Gray H. Twombly* 311
The Estrin-Deprivation Theory of Menstruation. *C. F. Fluhmann* 318
Study of Ovaries after Administration of a Gonadotropic Principle of Anterior Pituitary. *E. C. Hamblen* 321
Effect of Estrogenic Substance on Blood Volume. *Mae Friedlander, Norman Laskey and Samuel Silbert* 329
Migraine and Ovarian Deficiency. *S. J. Glass* 333
The Effect of Injection of Residual Ovarian Extracts. *H. W. Marlow* 339
The Biologic Effects of Thymectomy. *Nathan H. Einhorn and Leonard G. Rowntree* 342
The Biological Effects of Pineal Extract (Hanson). *L. G. Rowntree, J. H. Clark, A. Steinberg and A. M. Hanson* 348
An Iodine Tolerance Test for the Investigation of Thyroid Function. *E. M. Watson* 358
The Presence of an Excess of 'Male' (Comb-Growth and Prostate-Stimulating) Hormone in Virilism and Pseudohermaphroditism. *S. Levy Simpson, P. de Fremery and Alison MacBeth* 363
Arrhenoblastoma Case Report. *L. Grant Baldwin and James A. Gafford, Jr.* 373
Atrophy of the Adrenals and Addison's Disease. *William Susman* 383
Prostatic Atrophy Following Testicular Feeding. *Harold B. Myers, Ben Vidgoff, George H. Schade and Warren C. Hunter* 389
Clinical and Laboratory Notes
A Note on the Condition of the Endocrine Glands in Myopathy of Nutritional Origin. *Sergius Morgulis and Howard C. Spencer* 393
The Extraction of Both the Gonadotropic and (Free or Total) Estrogenic Hormones from a Single Urine Sample. *S. C. Freed and O. Hechter* 396
Scientific Program of the Twentieth Annual Meeting of the Association 398
Current Endocrine Literature 400

NO. 4, JULY, 1936

The Treatment of Diabetes Mellitus with Insoluble Insulin Compounds. *P. A. Gray* ... 461
The Effect of Oestrogenic Hormone on Experimental Pancreatic Diabetes in the Monkey. *Warren O. Nelson and Milton D. Overholser* 473
The Effect of Complete and Partial Hypophysectomy in Adult Albino Rats on Nitrogen, Calcium and Phosphorus Metabolism. *David Perla and Marta Sandberg* 481
The Composition of Weight Lost and the Nitrogen Partition of Tissues in Rats after Hypophysectomy. *Milton Lee and Gilbert B. Ayres* 489
The Effect of Hypophysectomy and of Phyone Injections on the Pancreas and Liver of the Newt. *A. Elizabeth Adams and Elsie N. Ward* 496
Quantitative Studies on the Structural Changes Induced in the Anterior Hypophysis by Injections of Oestrin. *J. M. Wolfe and C. S. Chadwick* 503
The Relationship of the Anterior Pituitary Gland to Thyroid and Ovary. *Leo Loeb, John Saxton and S. J. Hayward* 511
Assay of Blood and Urine for Thyreotropic Hormone in Thyrotoxicosis and Myxedema. *Saul Hertz and Eric G. Oastler* 520
Similarity of the Iodin Remission in Experimental Anterior Hypophyseal Hyperthyroidism, the Hyperthyroidism of Acromegaly and that of Exophthalmic Goiter. *Harry B. Friedgood* 526
The Influence of Pregnancy on Resistance to Thyroxine, with Data on the Creatine Content of the Maternal and Fetal Myocardium. *M. Bodansky and Virginia B. Duff* ... 537
Age as a Factor in the Resistance of the Albino Rat to Thyroxine, with Further Observations on the Creatine Content of Tissues in Experimental Hyperthyroidism. *M. Bodansky and Virginia B. Duff* 541
Analyses of Urine of the Chimpanzee for Estrogenic Content during Various Stages of the Menstrual Cycle. *Edgar Allen, A. W. Diddle, T. H. Burford, and J. H. Elder* 546
The Rôle of the Corpora Lutea in Prolonging the Life of Adrenalectomized Rats. *Frederick E. Emery and Edward L. Schwabe* 550
The Influence of Environmental Temperature and Salts upon the Survival Period of Adrenalectomized Rats. *Russell S. Weiser and Earl R. Norris* 556
Clinical and Laboratory Notes
An Investigation of the Presence of Estrogenic Hormones in the Ova and Ovaries of Fish. *Abner I. Weisman, Christopher W. Coates and Rosalind L. Moses* 561

The Isolation from Ovarian Tissue of a Crystalline Substance Possessing High Estrogenic Properties. *Robert H. Andrew and Frederic Fenger* 563
Weight Changes Following the Use of Glycerin Extract of Adrenal Cortex. *R. G. Hoskins and H. Freeman* 565
A Case of Adrenal Cortical Tumor in an Adult Male Causing Gynecomastia and Lactation. *H. Lisser* 567
Current Endocrine Literature 570

NO. 5, SEPTEMBER, 1936

Childhood Myxedema. *Charles Gilmore Kerley* 611
The Impedance Angle and Thyroid Disease. *M. A. B. Brazier* 621
Creatine Studies in Thyroid Disorders. *George W. Thorn* 628
Effect of Total Thyroidectomy upon Experimentally Induced Menstruation in Mature Ovariectomized Monkeys. *T. H. Burford, Edgar Allen and A. W. Diddle* 635
A Modification of the Frank-Goldberger Blood Estrin Test. *Theodore Neustaedter* 639
Tube-Locking of Ova by Oestrogenic Substances. *Rae Whitney and H. O. Burdick* 643
The Metabolism of Levulose: IX. The Influence of Endocrine Dysfunction on Tolerance. *Allen Winter Rowe, Mary A. McManus, Albert J. Plummer and Gertrude A. Riley* .. 648
The Spontaneous Activity of Adrenalectomized Rats Treated with Replacement and Other Therapy. *Curt P. Richter* 657
Fundamental Factors in the Interpretation of Stimuli Influencing Endocrine Glands. *H. Selye and J. B. Collip* 667
The Relation of the Basophilic Cells of the Human Hypophysis to Blood Pressure. *A. T. Rasmussen* 673
Clinical and Laboratory Notes
The Pituitary-Ovarian Relationship in a Human Hypophysectomized Female. *C. A. Elden* 679
Relative Effectiveness of Progestin in Hypophysectomized and Normal Rabbits. *Samuel R. M. Reynolds, Warfield M. Firor and Willard M. Allen* 681
Secretin XI: Its Influence on the Reticulocytes of the Circulating Blood. *John Ferguson* 683
Mortality Following Bilateral Adrenalectomy Combined with Renal Decapsulation. *M. Bodansky, C. M. Agress, Katherine R. Campbell and Cornelius L. Herrmann* 684
Relation Between the Occurrence of Endemic Goiter and the Presence of Traces of Silver and Barium in Drinking Water. *C. H. Boissevain and W. F. Drea* 686
Current Endocrine Literature 688

NO. 6, NOVEMBER, 1936

The Human Corpus Luteum and Progestin, Studies II. *J. P. Pratt, E. C. Hamblen, O. Kamm and D. A. McGinty* 741
The Effect of Estrogenic Substances upon the Pituitary, Adrenals and Ovaries. *Eugene T. Ellison and John C. Burch* 746
The Time Element in the Pituitary-Ovarian Response to Large Doses of the Estrogenic Hormone. *Charles Mazer, S. Leon Israel and Bernard J. Alpers* 753
The Effect of Interruption of the Supraoptico-Hypophyseal Tracts on the Antidiuretic, Pressor and Oxytocic Activity of the Posterior Lobe of the Hypophysis. *C. Fisher and W. R. Ingram* 762
The Endometrium in "Endometrial Hyperplasia" after Therapy. *E. C. Hamblen* 769
Treatment of Sexual Underdevelopment in the Human Male with the Anterior-Pituitary-Like Hormone (A.P.L.) of Pregnancy Urine. *Daniel L. Sexton* 781
Visceral Temperatures in the Intact and Unanaesthetized Animal. II. The Uterus of the Rat. *James B. Hamilton* 788
Extracts Containing Cortin. *Frank A. Hartman and W. D. Pohle* 795
The Genesis of Thyroid Protein: Clinical Assays of Artificial Thyroid Protein in Human Myxedema. *W. T. Salter and J. Lerman* 801
The Influence of the Intake of Calcium on the Blood Iodine Level. *Juanita Thompson* .. 809
Subclinical Hypothyroidism in Children. *Matthew Molitch and Sam Poliakoff* 816
Nitrogen and Creatine Metabolism in Relation to Environmental Temperature and Thyroid Function. *M. Bodansky and Virginia B. Duff* 822
The Significance of Electrical Impedance Measurements on the Human Body. *J. W. Horton and Saul Hertz* 831
Case Reports
Thyroid Dysfunction in Hemochromatosis. *M. Norman Orgel and David Barr* 839
Fugitive Acromegaly. Acromegaly Followed by Pituitary Cachexia. Treatment with an Alkaline Extract of the Anterior Pituitary. *E. Kost Shelton, Lyman A. Cavanaugh and Paul R. Patek* 846
Hypertrophy of the Testes with Symptoms of Hyperorchidism. *J. K. Fancher* 852
The Treatment of Pituitary Infantilism with Anterior Pituitary Extract. *Max M. Goldberg* 854

Pituitary Infantilism with Diabetes Mellitus. *Daniel L. Sexton and Fritz Neuhoff* .. 856
Clinical and Laboratory Notes
A Simple Method of Hypophysectomy in the Dog. Richard J. Bennett, Jr. 860
On the Evaluation of the Potency of Estrogenic Substances. *S. C. Freed and S. Soskin* 863
Further Investigation of the Hormone Content of Saliva Using the Female Bitterling Test. *Abner I. Weisman* 864
Dating Endocrine Substances: A Plea for Fresh Products. *Israel Bram* 866
Current Endocrine Literature 867
Index to Volume 20
Original Articles 881
Abstracts 886

ENDOCRINOLOGY

THE BULLETIN OF
THE ASSOCIATION FOR THE STUDY
OF INTERNAL SECRETIONS

VOLUME 20 JANUARY, 1936 NUMBER 1

ACTION OF ANTERIOR PITUITARY HORMONES ON BASAL METABOLISM OF NORMAL AND HYPOPHYSECTOMIZED PIGEONS AND ON A PARADOXICAL INFLUENCE OF TEMPERATURE[1]

OSCAR RIDDLE, GUINEVERE C. SMITH, ROBERT W. BATES, CLARENCE S. MORAN AND ERNEST L. LAHR

Carnegie Institution of Washington, Station for Experimental Evolution

COLD SPRING HARBOR, NEW YORK

INTRODUCTION

Two years ago it seemed desirable and possible to undertake a fairly comprehensive study of the action of various anterior lobe hormones on basal heat production. Such a study obviously required two quite distinct lines of investigation. First, the assay of all preparations for their content of some or all of the various pituitary hormones. Second, the determination in normal, hypophysectomized and thyroidectomized animals of the action of these attested preparations on the rate of heat production. This laboratory was then in favorable position to undertake this bi-partite study.

Most American and some European laboratories engaged in pituitary study have generously supplied us with preparations which they had assayed for one or another response and upon which, in many cases, much physiological study had been made. We consider all those who have thus shared their preparations as collaborators in this study. Though we have made, tested and variously studied many preparations of our own, a necessarily wide survey of the subject was possible only with the coöperation of all those individuals or laboratories named in our tabulated data.

MATERIAL AND METHODS

The first section of the study was carried out on immature male ring doves—whose crop-glands provide a quantitative assay of prolactin, whose testes measure the amount of true follicle-stimulating hormone (F.S.H.), and whose thyroids roughly measure the thyreotropic hormone. Our tests for F.S.H. and thyreotropic hormones are not yet sufficiently standardized

[1] Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 10, 1935.

This work was aided by a grant from the Carnegie Corporation of New York.

to permit us to translate the observed changes of weight into units; but our tests do permit us to state with fair certainty the (practically complete) absence of these hormones, to grade thyreotropic action, and roughly to quantitate F.S.H. potency. Moreover, our measure of F.S.H. potency is not subject to the wide error introduced by the "augmentation phenomenon" in assays made on rodent ovaries. Usually our measure of thyreotropic potency was based solely upon thyroid weight of treated in comparison with thyroid weight of untreated controls of the same race; in some cases, however, the apparent absence of thyreotropic, like its apparent presence, was checked by histological study.

All of our own preparations were made from pituitaries of cattle unless this is otherwise indicated in the tables. All F.S.H.+ preparations and all those of prolactin with numbers lower than 237 were made by the method originally described (1); later prolactin preparations were made by a modification of that method (2).

Since the preparations used in this study were assayed (by us) for only three hormones some of our readers who think the anterior lobe produces six or eight hormones may at once be tempted to conclude that we have not obtained sufficient information concerning any preparation to warrant its use in such a study of heat production as is made here. We ourselves shared more of such solicitude at the beginning of the study than we possess at its end, and the reader is asked momentarily to reserve judgment. If future work should show that only F.S.H., prolactin and thyreotropic—or these plus a very heat-labile hormone originally contained in our prolactin preparations—exist as separate entities in the pituitary, or if an additional group of hormones exist but are without effect on heat production, our conclusions concerning heat production will later require little or no revision. Finally, as already noted, many of the preparations used here have been studied for certain properties by others; the union of the earlier with present results may throw better light upon the real hormonal basis of several known responses to anterior pituitary dosage, and thus provide indications that the actual number of A.P. hormones is much less than the alleged number.

The second part of the study was made on adult normal ring doves and normal pigeons; and on a group of completely hypophysectomized, a smaller group of incompletely hypophysectomized, and on a single completely thyroidectomized pigeon. Measurements were made at night in a Benedict multiple-chamber closed-circuit apparatus on birds previously injected with hormone from 2 to 9 days. The birds were fasted for 24 hours—and held during this fasting period in a large glass cage at the same temperature at which their heat production[2] was later to be measured. Two to 6 birds were used in each test. The test involved measurement of each bird before injection (control) and usually at 2 points (on third and ninth days) within the period of injection. To avoid missing a possible time-restricted response, however, some tests were made at 2, 4 and 6 days after the first of the series of daily (or twice daily) injections; also in the case of a few pituitary preparations the

[2] On the curves "Calories" for doves are per 150 gram bird, per 4 hours; Calories for pigeons are per 300 gram bird, per 4 hours.

period from 2 to 19 hours after a single injection was studied, but with insignificant results which require no further consideration. Finally, and of much importance, the several heat production measurements outlined above were made at 2, or at 3, temperatures. In all cases measurement was made at

TABLE 1. RESULTS, FROM ORGAN WEIGHTS OF IMMATURE RING DOVES, OF ASSAYS OF VARIOUS A.P. PREPARATIONS FOR PROLACTIN, F.S.H. AND THYREOTROPIC HORMONES

Preparation and No.		Indications on quantity or presence of the three hormones			
		Units of prolactin		Thyreotropic	F.S.H.
		Per mg.	Per cc.		
Whole A.P. (beef) extract	156		5	++	I?
Whole A.P. (beef) extract	251		2	++	II
Whole A.P. (sheep) extract	172		15	++	II
Phyone*	41A		35	++	III
"	41B		3	++	II
Antuitrin G[1]	778-A		7	++	0
Squibb's Growth H.[2]	15997		0.5	++	II
Collip's Growth H.[3]	Q16		0.5	+	0
" " "	Q40A		0.5	+	0
Own Growth (Collip's meth.)	337	1	20	?	0
Prolactin (from beef)	58	0.5		0	0
"	74	0.5		+	0
"	157A	5		0	0
"	165	4		?	0
"	205	3		+	0
"	212	3		0	0
"	237	2		+	0
"	237b	1		0	0
"	256	4		0	0
"	256b	3		0	0
"	261	3		0	0
"	261b	1		0	0
"	308	1		0	0
"	308b	0.5		0	0
"	324	3		0	0
"	327A	4		0	0
"	327Ah	1		0	0
"	380h	8		0	0

* Cooperation of Dr. David Klein, Wilson Laboratories, Chicago.
1 Cooperation of Dr. Oliver Kamm, Parke, Davis and Company.
2 Cooperation of Dr. J. A. Morrell, E. R. Squibb and Sons.
3 Cooperation of Prof. J. B. Collip, McGill University, Montreal.

30° C.—earlier shown in this laboratory and elsewhere to be the "critical" temperature, or "zone of thermal neutrality" for the pigeon—and again at 15° C. or at 20° C. or at both of these lower temperatures.

RESULTS

In tables 1 and 2 are listed all of the preparations used in this study of heat production, together with a condensed indication of the amount of prolactin, thyreotropic and follicle-stimulating hormone found in each preparation. Altogether 47 different preparations were used, assays of each preparation for

three hormones being made on a group of 2-10 immature doves or pigeons. Obviously the details of these tests are beyond possibility of inclusion in this paper, and the reader must be asked to accept the summaries of tables 1 and 2 as the results of those numerous assays. The curves of figures 1-5 indicate whether and to what extent each preparation modified O_2-consumption. Since

TABLE 2. RESULTS FROM ORGAN WEIGHTS OF IMMATURE RING DOVES, OF ASSAYS OF A.P. PREPARATIONS FOR PROLACTIN, F.S.H. AND THYREOTROPIC HORMONES

Preparation or hormone and No.		Indications on quantity or presence of the three hormones			
		Units of prolactin		Thyreotropic	F.S.H.
		Per mg.	Per cc.		
Thyreotropic, Schering*		0.1		++	II
Thyreotropic, Winthrop[1]			0?	+++	II
Loeb's acid extract[2]			0.5	++	II?
Collip's thyreotropic[3]	TD2		0.2	++	I
" "	TG		0.5	++	II
Desiccated thyroid, fed		0		—	—
F.S.H.+(our own) pig	126	0.01		++	III
" beef	139	0.01		++	IV
" sheep	141	0.06		+	II
" pig	198	0.05		+++	IV
" beef	243	0.07		++	IV
" beef	243b	0.05		++	III
" pig	267	0		+++	III
Mare serum[4]			tr.?	0	III?
Luteinizing[5]	11R3	0.5		+	III
Adrenotropic (Own)	340		5	?	0
Prolan, Elberfeld[6]		tr.?		0?	I
Prolan (*special* Antuitrin S)[6]	315-E	tr.?		0	V
Estrin (amniotin)[7]			0	0	0

* Cooperation of Dr. Erwin Schwenk, Schering Corporation, and Schering-Kahlbaum Company.
1 Cooperation of Dr. John W. Hart, Winthrop Chemical Company, and the I. G. Farbenindustrie Aktiengesellsch., Elberfeld, Germany.
2 Cooperation of Dr. David Marine, Montefiore Hospital.
3 Cooperation of Prof. J. B. Collip, McGill University, Montreal.
4 Cooperation of Dr. Oliver Kamm, Parke, Davis and Company.
5 Cooperation of Prof. F. L. Hisaw and H. L. Fevold, University of Wisconsin.
6 Cooperation of Dr. F. Laquer, I. G. Farbenindustrie Aktiengesellsch. Elberfeld, Germany.
7 Cooperation of Dr. J. A. Morrell, E. R. Squibb and Sons.

100 curves—each presenting heat production averages obtained from 2-6 adult birds—are presented they obviously can not be considered individually here.

Whole A.P. extracts contain all three hormones (table 1) and in measurements made at 30° C. they greatly increase the heat production of normal doves (fig. 1) and hypophysectomized pigeons (fig. 5). At 15° C. these extracts decrease the heat production of normal doves (fig. 1), and at 20° they have little or no effect (2 curves, fig. 2). The F.S.H. content of these extracts is negligible here, since that hormone will be shown to have no effect

on heat production. Prolactin and thyreotropic are present in large amount in these extracts and are therefore possible sources of the very great calorigenic effect. Control heat production values are in each case given at the extreme left.

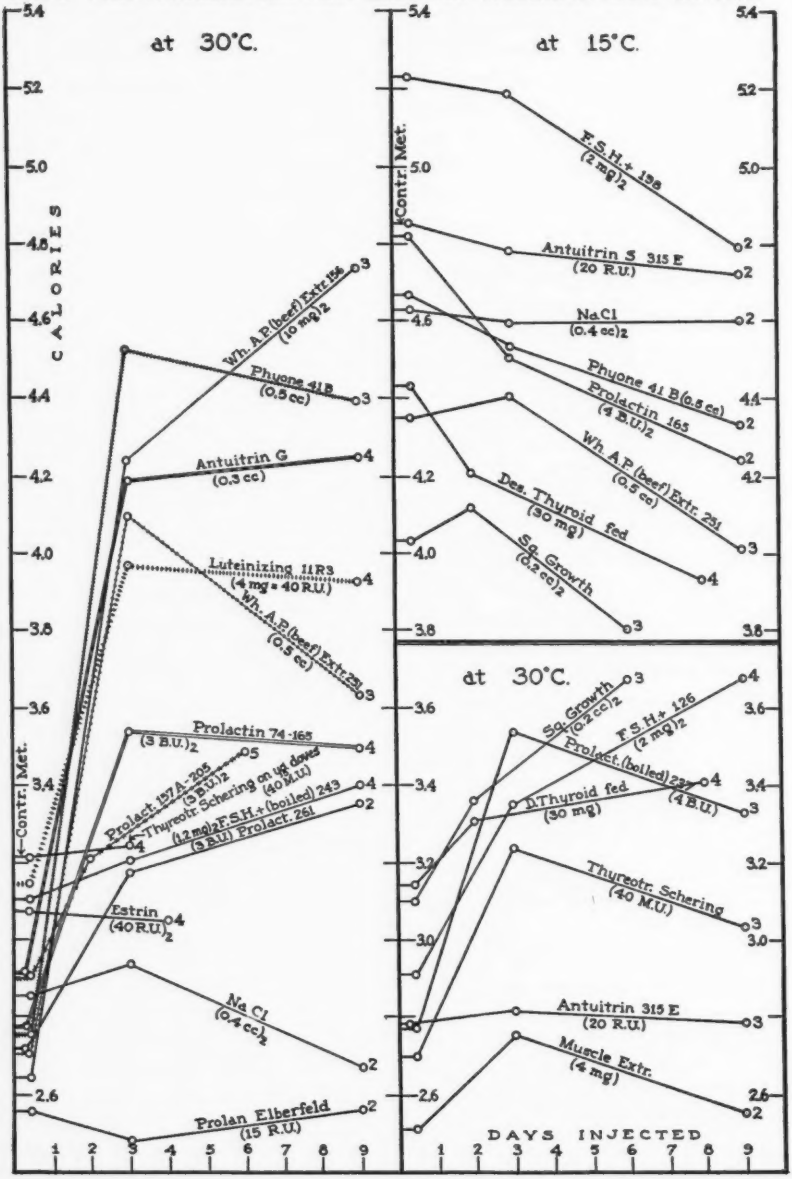
We here explain that the amount of daily dosage is stated on the curves in all cases; when the daily dose was administered in two injections the figure "2" is subjoined to indicate that fact. The digits at the free end of a curve indicate the number of birds used in the test. The reader will observe that the "control" values obtained for various similar groups (e.g., ring doves) may scatter rather widely. In explanation we note that much of this scatter arises from the fact that different races of doves compose the different groups, and that groups were measured at all seasons. Both race and season have earlier been shown to have marked effects on heat production in doves and pigeons (3).

The various growth hormone preparations are all found (table 1) to contain some or much prolactin and thyreotropic. Both of those of Collip, and a recent (1935) preparation of Antuitrin G (Parke, Davis and Company), are quite free from F.S.H. A preparation of the Evans growth hormone, made by us by the Evans 1931 method (and not tested on B.M.R.), contained the three hormones in approximately the proportions found in Phyone 41B. Some of the earlier growth preparations of Lee and Schaffer were assayed for hormone content—a very small amount of prolactin and much F.S.H. and thyreotropic being found—but their action on heat production was not studied by us. The growth preparations (Phyone 41A; Antuitrin G; and our own No. 337) which at 30° most strongly increased calorigenesis in normal (figs. 1, 3), hypophysectomized (fig. 4 and at 20°, fig. 5) and thyroidectomized (fig. 5) birds were the growth preparations which contained most prolactin and/or thyreotropic. The Squibb preparation, quite low in prolactin and strong in thyreotropic, had less effect on the B.M.R. (fig. 1) than preparations (Phyone 41B; Antuitrin G) which had equal thyreotropic and much more prolactin. Unfortunately, Collip's growth preparations were tested solely at 20° C. (fig. 2) before we had learned of the dominating rôle of temperature on the calorigenic action of these hormones.

Three hypophysectomized pigeons and one normal pigeon were measured at 14° C. after 3 and 8 days' dosage with 0.5 cc. of the growth preparation, Antuitrin G. None of the 5 figures prepared can accommodate data obtained at this temperature and the results require description. During dosage the average gain in body weight of the 4 birds was 17 per cent. At 14° the 3 operated birds showed a 7 per cent decrease in heat production after 3 days dosage, and at 8 days a return to the normal 14° level. Actual values at control and later measurements, in the order mentioned were 4.80, 4.44, and 4.77 Calories per kilo per hour. At 14° the normal pigeon's heat production was decreased at 3 days by 5 per cent and at 8 days by 45 per cent. At the 8-day test and at 14° the heat production of this normal pigeon was only 2.94 though its uninjected control value at 14° C. was 5.30 Calories per kilo per hour.

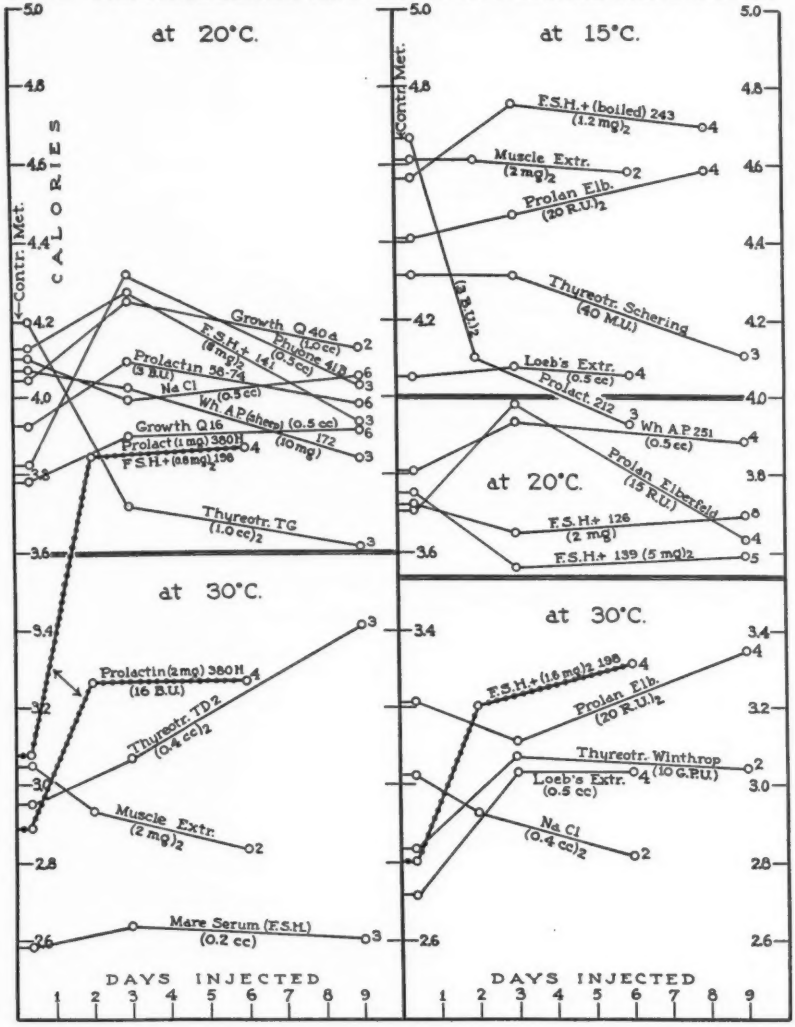
Most of the 18 prolactin preparations used (table 1) tested free of both thyreotropic and F.S.H.; but 4 of these could not be declared free of thyre-

FIG. 1 A.P. HORMONES AND B.M.R. OF NORMAL RING DOVES


at 30°C.
at 15°C.
at 30°C.
CALORIES
Contr. Met.
DAYS INJECTED
Wh. A.P.(beef) Extr. 156 (10 mg)2
Phyone 41B (0.5 cc)
Antuitrin G (0.3 cc)
Luteinizing HRS (4 mg = 40 R.U.)
Wh. A.P.(beef) Extr. 251 (0.5 cc)
Prolactin 74-165 (3 B.U.)2
Prolact. 157 A-205 (3 B.U.)2
Thyreotr. Schering on 1/8 doses (40 M.U.) (1.2 mg) F.S.H.+(boiled) 243 (3 B.U.) Prolact. 251
Estrin (40 R.U.)2
Na Cl (0.4 cc)2
Prolan Elberfeld (15 R.U.)
F.S.H.+ 198 (2 mg)2
Antuitrin S 315 E (20 R.U.)
Na Cl (0.4 cc)2
Phyone 41 B (0.5 cc)
Prolactin 165 (4 B.U.)2
Wh. A.P. (beef) Extr. 251 (0.5 cc)
Des. Thyroid fed (30 mg)
Sq. Growth (0.2 cc)2
Sq. Growth (0.2 cc)2
F.S.H.+ 126 (2 mg)2
Prolact. (boiled) 251 (4 B.U.)
D. Thyroid fed (30 mg)
Thyreotr. Schering (40 M.U.)
Antuitrin 315 E (20 R.U.)
Muscle Extr. (4 mg)

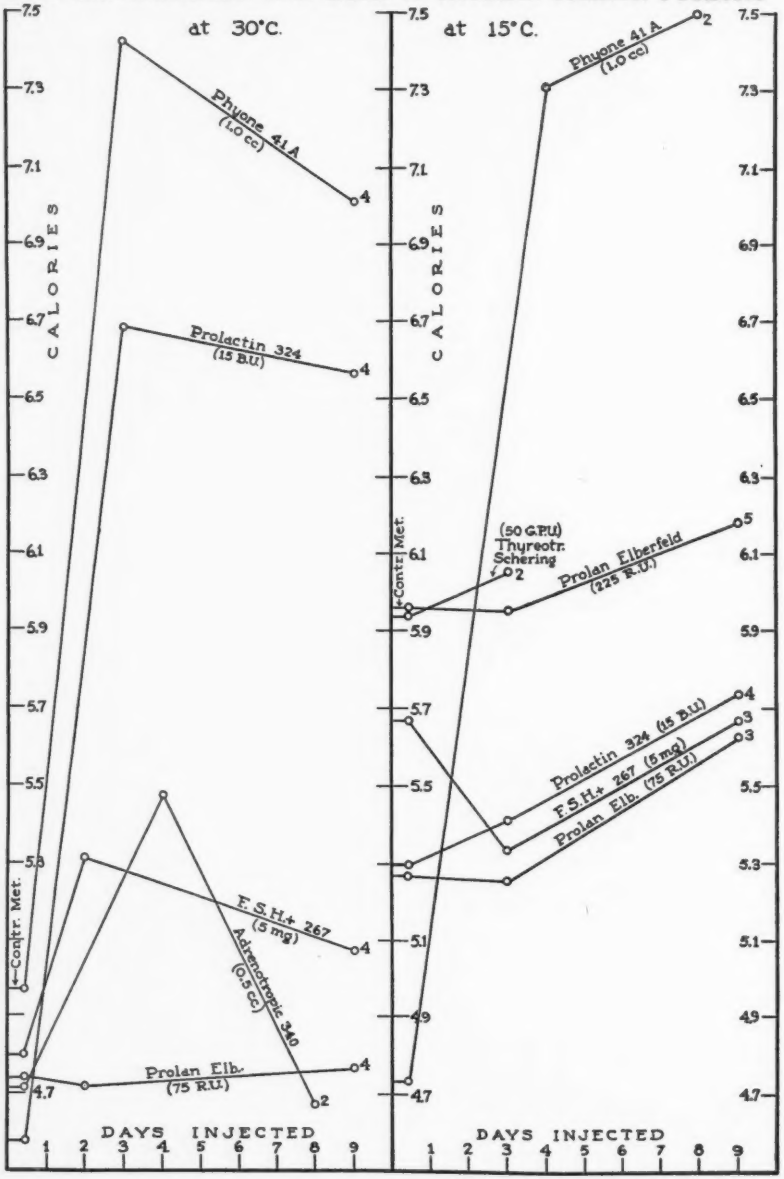
otropic. Six heated prolactin preparations were used—4 of them having been heated to boiling for 1 hour, 2 others heated to 60° C. for 5 hours. At 30° C. 10 curves (figs. 1-5) indicate an increased heat production in each case,

FIG. 2 A.P. HORMONES AND B.M.R. OF NORMAL RING DOVES



although very low dosage of 3 partially hypophysectomized doves with No. 308 (fig. 5) gave only an insignificant (6 per cent) increase. At 30° normal doves and pigeons, and hypophysectomized and thyroidectomized groups, are all found to respond to prolactin—heated and unheated—by a marked rise in heat production. In general, this increase is greater at higher dosage levels. At 15° prolactin decreased the metabolism in normal doves (figs. 1, 2) and insignificantly affected it in normal common pigeons (fig. 3). At 20° low dosage with prolactin had no effect in normal doves (fig. 2); in pigeons partially and completely hypophysectomized (3 curves) it moderately increased the metabolism (fig. 5).

FIG. 3. A.P. HORMONES AND B.M.R. OF NORMAL COMMON PIGEONS



The thyreotropic preparations used contained practically negligible traces of prolactin (table 2), and even traces could not be declared in the very potent German product obtained through the Winthrop Chemical Co. All

thyreotropic preparations contained F.S.H.—usually in very fair quantity. All these preparations, excepting only the Schering-Kahlbaum product, were obtained in solution and this of course introduces an element of uncertainty that does not attach to the assay of powdered preparations; but this uncertainty attaches merely to questions of concentration and as to whether the original solutions had more potency—for either or all of the 3 hormones—than was found and tabulated by us.

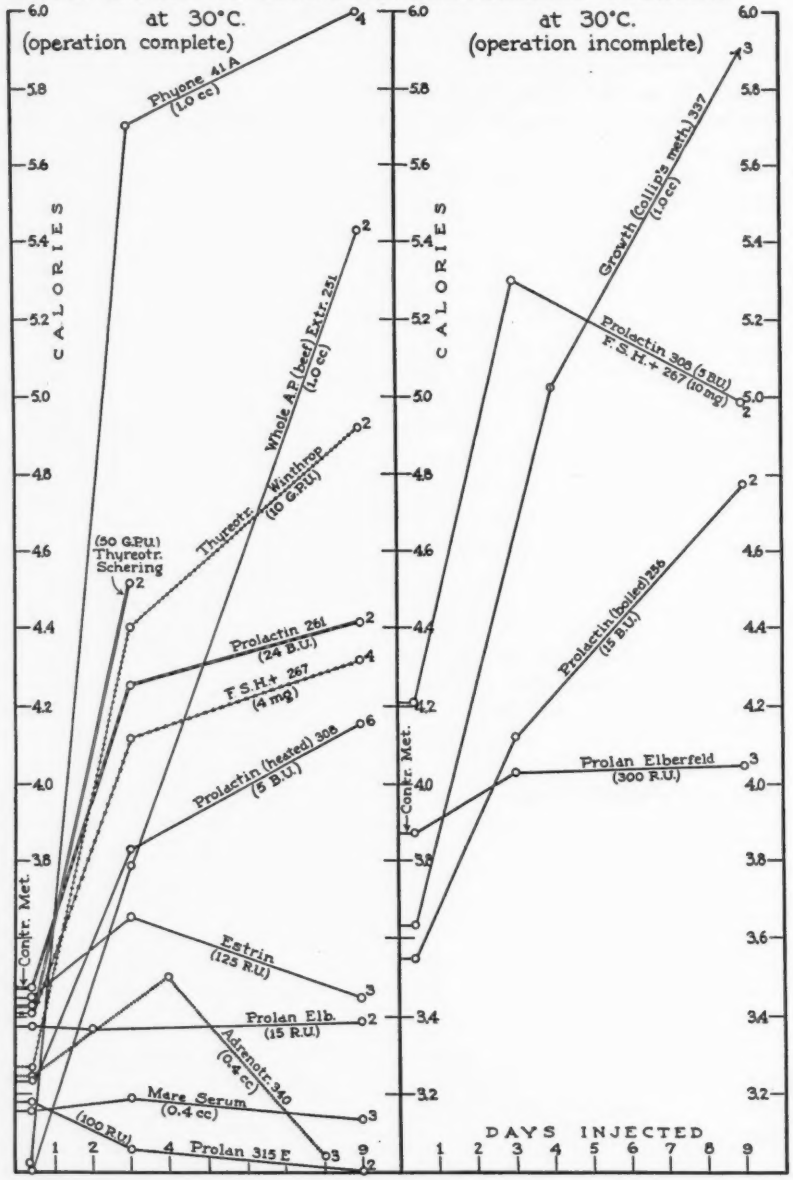
In 30° measurements the Schering thyreotropic increased the B.M.R. of adult normal ring doves, but not (3 day test only) in juvenile ring doves whose control B.M.R. was notably higher than that of the adults (fig. 1); at 15° C. the preparation apparently decreased heat production in normal adult ring doves (fig. 2), and had no effect (fig. 3) in normal common pigeons (dosage on figs. 1 and 2 should read G.P.U., not M.U.). In hypophysectomized pigeons it markedly increased heat production at 30° (fig. 4) and at 20° C. (fig. 5); in a thyroidectomized pigeon at 30° it caused no significant change in rate of heat production.The Winthrop thyreotropic at 30° gave only a slight increase in normal doves (fig. 2) but very marked increases in hypophysectomized pigeons—both at 30° (fig. 1) and at 20° C. Loeb's acetic acid extract in ring doves produced a moderate increase at 30° and was entirely without effect at 15° C. (fig. 2). At 15° this extract gives a curve which is indistinguishable from its "muscle extract" control. Used on normal ring doves Collip's thyreotropic preparations (TD2 and TG) moderately increased heat production at 30°, and at 20° definitely decreased heat production (fig. 2). Feeding of desiccated thyroid (30 mg. daily) to ring doves (150 gram body weight) produced effects similar to those obtained from thyreotropic hormone —increased heat production at 30° and a definite decrease at 15° C. (fig. 1).

Our preparations designated "F.S.H.+" usually have a very high concentration of the true follicle-stimulating hormone, a high concentration of thyreotropic, and are nearly or quite free from detectable prolactin (table 2). Since F.S.H. itself will be found wholly without effect, the thyreotropic content of the best of these preparations (126, 139, 267) may be expected to account for the observed changes in heat production. The present study (see below) suggests, however, that the amount of prolactin still remaining in preparations 141, 198, 243 and 243b is not negligible at higher dosage levels (5-10 mg.). For most of such highest dosage it will be observed that the best (No. 267) of the listed preparations was used.

These F.S.H.+ preparations, used in 30° tests, increased calorigenesis in normal doves (figs. 1, 2), in normal pigeons (fig. 3) and in incompletely hypophysectomized pigeons (fig. 4). At 15° these preparations decreased heat production in normal doves (fig. 1) or left it unchanged (fig. 2); in normal pigeons it was unchanged (fig. 3). At 20° there was no change (3 curves) in normal doves (fig. 2).

In the estrin (Amniotin, Squibb) preparation used we could find no trace of F.S.H., prolactin or thyreotropic hormone (table 2). In normal doves 40 R.U. daily had no effect at 30° in a 4-day test (fig. 1). In hypophysectomized pigeons measured at 30° a daily dosage of 125 R.U. probably produced no change in heat production at 3 and 9 days (fig. 5). These few tests with

FIG. 4. B.M.R. OF HYPOPHYSECTOMIZED PIGEONS



estrin were made to learn whether in female pigeons (with large oviducts) the estrin produced as a secondary response to F.S.H. administration may be expected to cause a measurable increase in heat production.

Mare serum has high F.S.H. potency, no detectable thyreotropic, and possibly or questionably a trace of prolactin (table 2). At 30° it had no effect on heat production of normal doves (fig. 2), hypophysectomized doves (fig. 5) and hypophysectomized pigeons (fig. 4). At 20° it probably had no effect on partially hypophysectomized pigeons (fig. 5). We consider these tests with very potent mare serum as decisive evidence that F.S.H. has no effect on heat-production.

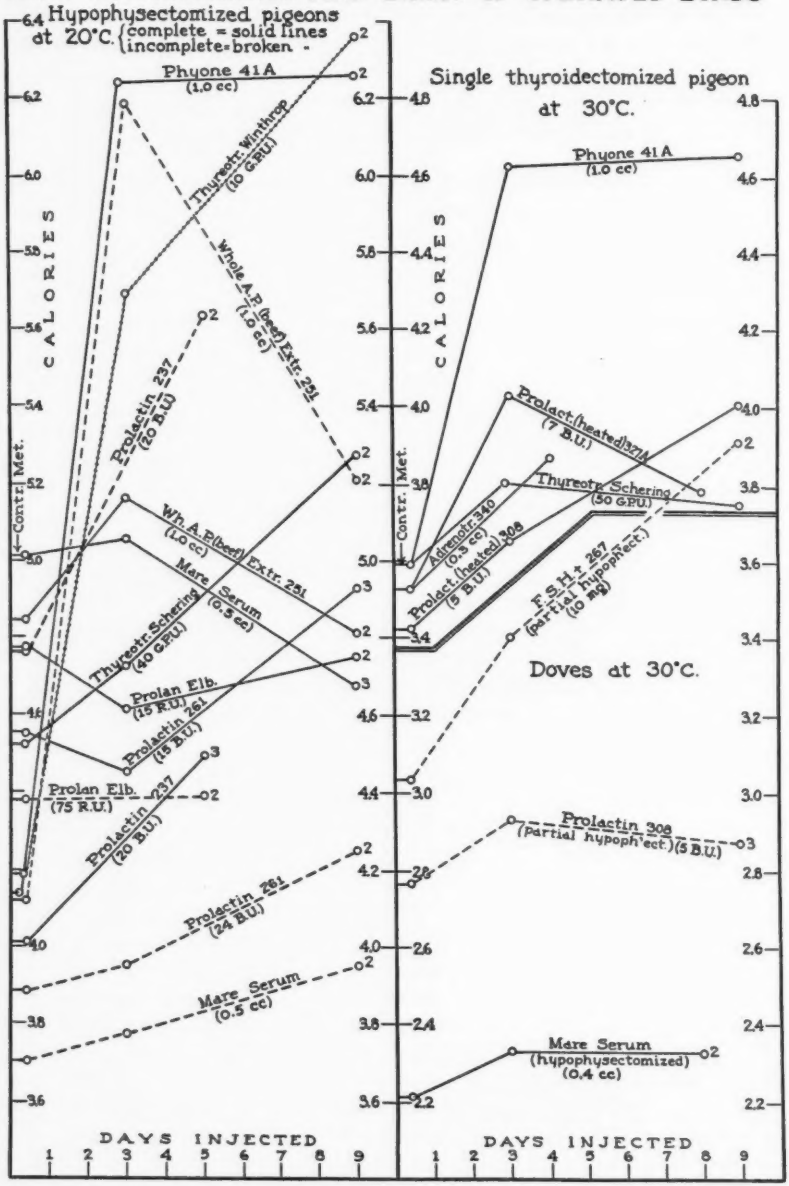
Prolan of 2 types were used (table 2). Our assays of the Elberfeld product indicated true F.S.H. potency of rather low degree (the immature pigeon testis is not subject to the "augmentation phenomenon"), thyreotropic potency absent or questionable, and prolactin absent or present in traces only. Prolan (or Antuitrin S) No. 315-E was a special research preparation, supplied by Dr. Oliver Kamm, of intense F.S.H. potency (in rats, with "augmentation," = 1,000 R.U. per mg.), without thyreotropic or definitely detectable prolactin in the dosages used. Numerous tests (all kinds of birds at all temperatures) with these prolans show them to be without effect (confirming Reiss and Winter, 4—for rabbits; Gaebler, 5—for dogs) on heat production. Since this was learned early these preparations—containing 1 A.P. hormone—were often substituted for NaCl or "muscle extract" as controls for other birds whose heat production was being measured at the same time. These tests, and those made with mare serum, seem to prove definitely that F.S.H. has no calorigenic action whatsoever; and any "F.S.H." extract of pituitary origin which has calorigenic action probably contains thyreotropic hormone or prolactin. Unfortunately, it was not possible to obtain the Fevold-Hisaw F.S.H. in quantity permitting a study of its power to increase heat production.

With this demonstration of the inactivity of true F.S.H. on heat production in mind, and remembering the high thyreotropic content of our F.S.H.+ preparations, we may now examine the effect on heat production of combining thyreotropic and prolactin. Among the lowermost curves of figure 2, and to the right, is seen the effect at 2 and 6 days of injections of 1.6 mg. daily (in 2 doses) of preparation (F.S.H.+) No. 198 into a group of doves. Directly to the left is shown the effect of 2.0 mg. (once) daily injections of prolactin No. 380h (a heated preparation). Close to the latter curve is that obtained by using only one-half the previously used amounts of Nos. 198 and 380h, but using them in combination. Clearly, combined dosage with reduced amounts of the two hormones was more effective in increasing heat-production than was double that dosage of either hormone alone. Evidence of the same sort was obtained in the use of Phyone 41A (high prolactin and high thyreotropic, table 1) on a thyroidectomized pigeon (fig. 5). In this thyroidless bird, in which thyreotropic hormone proved ineffective, daily injections of 1.0 cc. of Phyone 41A produced at 30° less than half the increased heat production produced, *a*, at 30° in normal pigeons (fig. 3), *b*, at 20° in hypophysectomized pigeons (fig. 4) or *c*, even at 15° in normal pigeons (fig. 3).[3]

It is evident therefore that prolactin and thyreotropic exercise their heat-producing action on different tissues, and that a simultaneous action on these

[3] Later, thyreotropic was found to have no demonstrable effect in 2 additional thyroidectomized pigeons.

FIG. 5. A.P. HORMONES AND B.M.R. OF OPERATED BIRDS


Hypophysectomized pigeons at 20°C. {complete = solid lines incomplete=broken
Phyone 41A (1.0 cc)
Thyreotr. Winthrop (10 G.P.U.)
Whole A.P. (beef) Extr. 251 (1.0 cc)
Prolactin 237 (20 B.U.)
Wh. A.P. (beef) Extr. 251 (1.0 cc)
Mare Serum (0.5 cc)
Thyreotr. Schering (40 G.P.U.)
Prolan Elb. (15 R.U.)
Prolactin 261 (15 B.U.)
Prolan Elb. (75 R.U.)
Prolactin 237 (20 B.U.)
Prolactin 261 (24 B.U.)
Mare Serum (0.5 cc)
Contr. Met.
CALORIES
DAYS INJECTED
Single thyroidectomized pigeon at 30°C.
Phyone 41A (1.0 cc)
Prolact. (heated) 247A (7 B.U.)
Thyreotr. Schering (50 G.P.U.)
Adrenotr. 340 (0.5 cc)
Prolact. (heated) 308 (5 B.U.)
F.S.H. + 267 (partial hypophect.) (10 mg)
Doves at 30°C.
Prolactin 308 (partial hypophect.) (5 B.U.)
Mare Serum (hypophysectomized) (0.4 cc)
Contr. Met.
CALORIES
DAYS INJECTED

different tissues produces more than an additive effect on total heat production. This observation seems to throw much light on the very extraordinary capacity of all crude A.P. extracts to increase heat production—for they all contain

large amounts of both prolactin and thyreotropic. Growth hormone preparations are a kindred group since—though the calorigenically inert F.S.H. may be separated from them—they too contain more or less prolactin and thyreotropic, and in both normal and hypophysectomized birds they usually have relatively high calorigenic action. We therefore regard this as evidence that there is no individual or separate growth hormone, but that the familiar balanced growth response proceeds from the simultaneous action of more than one hormone—and that thyreotropic and prolactin are, or are among, those simultaneously acting hormones.

The assay of a recent (1935) luteinizing preparation of Fevold and Hisaw (table 2) showed it to be fairly high in true F.S.H. potency, to have low but definite thyreotropic content, and a moderate prolactin content. In normal doves at 30° 4 mg. daily of luteinizing (thyreotropic + prolactin) produced apparently the same increase (fig. 1) in heat production (25 per cent) that was obtained from 1 mg. daily of prolactin 380h (heated) combined with 0.8 mg. (in divided daily dose) of our thyreotropic containing preparation No. 198 (fig. 2). We do not attribute this heat producing activity to a unique (luteinizing) A.P. factor, but to the mixture of 2 of the 3 pituitary principles found in it.

A preparation here designated adrenotropic, made by us according to the method of Collip (6), was free from F.S.H., had questionable traces of thyreotropic, and fairly high prolactin potency (table 2). Three measures were made of its heat producing activity at 30° C. In normal pigeons a moderate increase at 4 days was followed by a decrease to normal at 8 days (fig. 3); in hypophysectomized pigeons a slight rise at 4 days was followed by a decrease to or below normal at 8 days (fig. 4); in a thyroidectomized pigeon there was a slight or moderate increase at 4 days, with no later measurement (fig. 5). If a separate adrenotropic hormone exists the amount of the preparation injected should have contained adequate amounts of that hormone. Reference to table 2 will show that the prolactin content of the amount injected daily in these common pigeons was only about 2 B.U.—a smaller quantity of prolactin than we ever injected as a purified prolactin preparation.

COMMENT

The consideration of several earlier studies in mammals on the relation of A.P. extracts to calorigenesis is here attended with difficulties; in each case a full consideration of the composition of the extract—often or usually unknown to us—would be required. We conclude that an attempt to review those numerous studies would require many pages and accomplish little that is constructive and wholly germane to this paper. The various studies of Loeb and Basset, Lee, Teel, and Gagnon, Siebert and Smith, Eitel and Loeser, Schittenhelm, Andersen and Collip, Szarka and Evans, Gaebler, Bueno and Barnes, and others, are well known. This earlier work dealing with heat production, together with still earlier work of Uhlenhuth, Smith, Riddle, Aron and Loeb on pituitary-thyroid relationships, mainly disclosed the existence of a thyreotropic hormone which, indirectly through the thyroid, increases heat production.

The present study shows that a second pituitary hormone, prolactin, has marked action on the rate of O_2-consumption in doves and pigeons—normal, hypophysectomized and thyroidectomized. Certain known facts would suggest that the suprarenals intermediate this action of prolactin, but the point or nature of the calorigenic action of prolactin is practically unknown.

The evidence here presented for an increased calorigenesis as a result of the simultaneous action of thyreotropic and prolactin is important. Though the basis or explanation of this result is quite unknown, it has been indicated that the two hormones have different sites or modes of action. Tentatively one may suppose that an unbalance of the two sources of heat stimulation (oxidation) is likely to bring limitations or restrictions on one or the other of these sources, and that such restrictions are absent when the two sources are concurrently active. Probably some factual support of this view is found in our observation that prolactin alone usually markedly increases body weight in doves and pigeons; prolactin plus thyreotropic slightly increases their body weight; thyreotropic (+ F.S.H.), with little or no prolactin, practically always rapidly decreases body weight.

Whatever the basis of the synergism to calorigenesis which results from concurrent injection of the two hormones the detection of the synergism has led to a more complete analysis of the materials of this study than would otherwise have been possible. Whole A.P. extracts which have remarkably high calorigenic action (at critical temperature, and during first few days) contain both of these two hormones. Assay of several preparations of "growth hormone" has shown in all cases the presence of some or much of both of these same hormones; and here too (in all preparations that have at least a moderate amount of each of the two hormones) this is associated with high calorigenesis. We have proved the presence of the two hormones in preparations repeatedly found by others to give a satisfactory growth response. Further, we observe that the combination of these same two hormones leads to an unusually high calorigenic response. We think these related facts throw doubt upon the individuality or separate existence of a "growth" hormone. In no case whatever, in our opinion, has it yet been proved that effects ascribed to a "growth" hormone were not joint effects produced by a mixture of two or more true and distinct A.P. hormones.[4]

Apparently of still greater consequence is the result obtained here on the effect of low temperature on the calorigenic action of both thyreotropic hormone and prolactin. Particularly in ring doves we find that these hormones increase heat production at 30°, and at 15° C. they tend to decrease it from the normal (high) 15° level. Since these hormones (and desiccated thyroid) must be considered normal endogenous agents of thermogenesis this effect of temperature is surprising, and seemingly incapable of acceptable explanation at present.

The drug, alpha-dinitrophenol, was earlier observed by Magne, Mayer and Plantefol (7) to have lost much of its calorigenic power in mice measured at

[4] Moreover, in the dwarf mouse thyreotropic and prolactin have likewise each been shown to cause some growth and to give synergism of growth when combined (Bates, Laanes and Riddle, Proc. Soc. Exper. Biol. & Med. Dec. 1935; in press).

9° to 11° C.; but this was not observed in dogs water-cooled at 14° C. Tainter (8) found that at 2° to 6° C. dinitrophenol failed to cause its usual metabolic stimulation in rats, guinea pigs and pigeons. In view of the results of the present study Riddle and Smith (9) made tests of the action of dinitrophenol in doves and pigeons at 30° and 15° C. They found that at 15° calorigenesis is notably reduced—though not abolished (or even reversed in doves) as was sometimes found for the prolactin and thyreotropic in the present study. Notable also was the observation that in a single thyroidectomized pigeon the dinitrophenol had relatively little calorigenic action at either 30° or 15° C.

It is therefore now evident that both endogenous and exogenous calorigenic substances are influenced by temperature in a paradoxical manner. At low temperatures—when increased heat apparently is needed—the injection of these substances results in a smaller increase (even no increase, or a decrease) than is obtained at a higher (critical) temperature. And, in many cases at least, the action of the calorigenic substance is decreased or abolished although the body temperature is not significantly decreased.

Since the "critical temperature," or the temperature of minimal heat production (30° in pigeons), is apparently also the temperature at which maximum calorigenic effects of these A.P. hormones may be obtained, it is obvious that temperature of measurement largely determines the extent to which a calorigenic A.P. extract will affect the rate of heat production. But metabolism studies in this field must deal with still another and even more embarrassing problem: If the critical temperature is the point, not only at which true "basals" must be obtained (we are not sure this is true), but at which A.P. extracts give their maximum calorigenesis, what is the critical temperature of the hypophysectomized animal—rat, dog, pigeon? Is it correct to compare the metabolism of a normal pigeon at 30° C. with that of the hypophysectomized pigeon at 30° C.? Elsewhere (10) we, like others, have made such a comparison; but we doubt its full validity. We think it improbable that the critical temperature, or "zone of thermal neutrality" is established in any individual (species) without the influence of its anterior pituitary. If that is true the removal of the pituitary should then shift the point of critical temperature up or down the thermometric scale. Would simultaneous removal of the posterior pituitary further shift the critical temperature? How would thyroid removal alone, either before or after hypophysectomy, influence the temperature of minimal heat production? These questions require answer before we can know, for example, whether there is validity or error in Houssay's (11) statement that the basal metabolism of thyroidectomized dogs is not further decreased by hypophysectomy.

SUMMARY AND CONCLUSIONS

Forty-seven anterior pituitary preparations, prepared in part by us and in part prepared and contributed to this study by other American and European laboratories, were assayed for prolactin, thyreotropic and follicle-stimulating hormone content on approximately 250 immature ring doves. Each of these assayed preparations was repeatedly tested for its action on heat production.

Many such tests were made on normal adult doves and on normal adult pigeons; other tests were made on hypophysectomized doves and pigeons; 5 such tests were made on a thyroidectomized pigeon. Heat production tests with all types of preparations were made on most types of pigeon and at three temperatures—30°, 20°, and 15° C.

The anterior pituitary contains not only one but two hormones which affect the rate of 0_2-consumption in all these types of birds. Prolactin has marked calorigenic action; unlike the thyreotropic hormone it does not act through the thyroid but in a manner now practically unknown.

Follicle-stimulating hormone and prolans have no calorigenic action whatever. A luteinizing preparation had calorigenic action which is fully explained by the amounts of thyreotropic and prolactin present in it. Tests of an adrenotropic preparation revealed some calorigenic action which is probably fully accounted for by its prolactin content.

The simultaneous injection of prolactin and thyreotropic produced a synergistic action on 0_2-consumption that is greater than the sum of their separate actions.

Various and typical preparations of "growth" hormone are found always to contain more or less of both calorigenic hormones—thyreotropic and prolactin. Growth preparations usually have very high calorigenic action—probably ascribable to the presence of the two calorigenic hormones—and the growth-promoting action of these preparations probably rests upon the joint and combined action of thyreotropic and prolactin. These results therefore throw doubt upon the existence of a separate or individual "growth" hormone in the anterior pituitary.

It is found that the calorigenic action of injected prolactin and thyreotropic hormone (or of feeding desiccated thyroid)—administered singly or in mixture—is influenced in a paradoxical manner by temperature. Preparations which greatly increase thermogenesis in normal and hypophysectomized animals at 30° C. usually have little or no effect on heat production at 20°, and some even decrease the normal 14° or 15° value when measurements are made on normal birds at this temperature.

REFERENCES

1. Riddle, O., R. W. Bates, and S. W. Dykshorn: Am. J. Physiol. **105:** 191. 1933.
2. Bates, R. W., and O. Riddle: J. Pharmacol. & Exper. Therap. **55:** 365. 1935.
3. Riddle, O., G. C. Smith, and F. G. Benedict: Am. J. Physiol. **107:** 333. 1934.
4. Reiss, M., and K. A. Winter: Endokrinologie **3:** 174. 1929.
5. Gaebler, O. H.: Endocrinology **19:** 63. 1935.
6. Collip, J. B.: J. Mt. Sinai Hosp. **1:** 28. 1934.
7. Magne, H. A., A. Mayer, and L. Plantefol: Ann. de physiol. **8:** 1. 1932.
8. Tainter, M. L.: J. Pharmacol. & Exper. Therap. **51:** 45. 1934.
9. Riddle, O., and G. C. Smith: J. Pharmacol. & Exper. Therap. **55:** 173. 1935.
10. Riddle, O., G. C. Smith, and C. S. Moran: Proc. Soc. Exper. Biol. & Med. **32:** 1610. 1935.
11. Houssay, B. A.: Endocrinology **18:** 409. 1934.

UTERINE RESPONSE TO PITOCIN[1]

MILO R. WHITE AND J. P. PRATT
Henry Ford Hospital
DETROIT, MICHIGAN

The effect of pituitrin on the pregnant uterus in the latter part of gestation is well known. In recent years the response of the uterus in its different physiologic phases to pituitrin and its oxytocic principle—pitocin has been studied experimentally and clinically. As a result of this more recent work, the reaction of the uterus in early pregnancy to posterior lobe extracts and especially to pitocin is now described as a means of diagnosing pregnancy and of differentiating the pregnant uterus from other abdominal or pelvic tumors. Since many of the experimental and clinical observations are not in entire agreement, it seemed worth while to undertake some studies of our own to test the merits of this method as a test for pregnancy in the human. In this paper we shall describe, for the most part, the response of the uterus in early pregnancy to the intravenous injection of pitocin and also reference will be made to the use of the method as a means of differential diagnosis.

In 1925 Hoehne-Greifswald (1) described his observations of the pregnant uterus following the intravenous injection of pituglandol, a posterior lobe extract. He reported that there was always a vigorous contraction of the uterus which became more pronounced as the irritability of the uterus increased due to the advancement of the pregnancy. An increase in the consistency following the injection of the oxytocic agent was observed as early as the second month of gestation and this reaction of the uterus was advocated as a means of differential diagnosis when the presence of a pregnancy was suspected.

Lorincz (2) in 1927 used intravenously a posterior lobe extract called pituisan and observed that the pregnant uterus contracted promptly and became ligneous in consistency. He also noted that the juncture between the body of the uterus and the cervix, which was hardly palpable before the injection, became easily palpable afterwards. The contraction of the uterus was found to persist for a period of five minutes, after which the musculature relaxed and became soft. The author reported that in sixty cases there were no untoward results so far as the patient and the pregnancy were concerned, although some of the women became pale, perspired freely and complained of nausea and paresthesias. Lorincz also found the test especially helpful in cases of retroflexion in which the enlarged cervix may easily be mistaken for the uterus and the retroflexed pregnant fundus may be misinterpreted as an adnexal tumor. In such cases the apparent separation between the fundus and the cervix promptly disappeared after the injection.

Zorn (3) has also advocated the use of this method as a means of determining whether an abdominal tumor is a pregnant uterus. This observer used 0.25 cc. of pituglandol intravenously in patients seven to ten months pregnant

[1] Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 11, 1935.

and noted that in thirty to sixty seconds there was a definite contraction of the uterus which persisted for one to five minutes. In no case was there any injury to the fetus, although in a number of instances the fetal heart rate was decreased. In five patients who were six weeks pregnant he was able to confirm the diagnosis of pregnancy by noting the increased firmness of the uterus after the intravenous injection of the oxytocic agent. Furthermore, he observed that a soft degenerating myoma did not show any increase in consistency after the injection.

Rodecurt (4) has used orasthin in his clinical work. This oxytocic agent is the same as pitocin and eliminates the vasopressor effect of pituitrin which is responsible for the unpleasant subjective symptoms, especially in patients with circulatory disturbances. In his experience the intravenous injection of orasthin has been a distinct aid in the differential diagnosis of pregnancy. Reeb (5) and Kustner (6) have also emphasized the value of this method in differentiating the pregnant uterus from other abdominal tumors. Reeb has pointed out that the Aschheim-Zondek test only indicates the presence or absence of pregnancy and is of no aid in differentiating the pregnant uterus from other abdominal tumors which may be present. In the experience of these observers the pregnancy is not jeopardized by the test.

The reaction of the uterus to pituitrin has been known for many years but the use of an oxytocic for the purpose of determining whether a pregnancy exists in the uterus has received almost no consideration in this country. Before one gives a physiological explanation for this alleged specific reaction of the pregnant uterus, one must first consider those factors already present which may affect the contractile motility of the uterus and its responsiveness to an oxytocic agent. While very little is known of the neurogenic control of uterine contractility, endocrinologic studies have contributed considerable information on humoral control. However, much of the experimental work is contradictory and that which has been done on animals may not be directly applicable to the human.

Knaus (7) in his studies on the rabbit found that the uterus in the presence of a corpus luteum loses its responsiveness to posterior lobe extract and that the contractility may be restored by extirpating the corpus luteum. That there is a close relationship between the corpus luteum and uterine motility in some animals is indicated by the fact that, in the rabbit, extirpation of the corpus luteum throughout pregnancy is almost invariably followed by abortion. There is, however, a distinct species variation in this reaction. Knaus (8) has also studied the response of the human uterus to pituitrin and found that the uterus becomes refractory to this oxytocic agent at the time of ovulation and therefore at the onset of corpus luteum formation.

Reynolds (9) has shown that uterine motility in rabbits is suppressed after castration and that the normal motility can be restored by the injection of theelin. Reynolds and Friedman (10) demonstrated a diminution in uterine motility in rabbits after coitus and in pseudo-pregnancy. In the non-pregnant rabbit they have been able to produce a diminution of uterine motility by injecting the urine of pregnant women. From this it may be inferred that the pregnant woman excretes in her urine a hormone which depresses the con-

tractile motility of the rabbit uterus either directly or secondary to changes induced in the ovaries.

From the foregoing experimental work and especially that of Knaus on the human uterus, it appears that one of the functions of the corpus luteum is to inhibit the uterine contractility so as to facilitate the nidation of the fertilized ovum. This inhibitory effect would in part account for the softening which is so characteristic of the pregnant uterus. If, as Fellner (11) and others have pointed out, one persists in the palpation of a softened pregnant uterus, the uterus gradually becomes smaller and much firmer in consistency. In the opinion of Fellner, the contraction of the pregnant uterus with palpation may so reduce its size as to appear as a hypoplastic organ, and the possibility of pregnancy will not receive serious consideration. This has been confirmed repeatedly in the course of our own observations that the uterus in early pregnancy will contract and become quite firm if it is palpated vigorously enough. While the corpus luteum may have an inhibitory effect on the contractility of the pregnant uterus as has been described by Knaus, the extent of inhibition is limited, for we have found that the gravid uterus contracts with palpation and shows an increase in consistency within one minute after the intravenous injection of 0.5 cc. of pitocin.

METHOD

In obtaining our clinical data, a routine pelvic examination was first made with special attention directed to the consistency of the uterus. The blood pressure and pulse were then taken, after which 0.5 cc. of pitocin was injected intravenously. One minute after the injection the pelvic examination was repeated to determine if any change in the consistency of the uterus had occurred. The examination was again repeated three minutes after the injection. The blood pressure and pulse were then taken and any subjective symptoms noted. In a number of cases the inability of the patient to relax properly, the thickness of the abdominal wall, and the position of the uterus made the evaluation of the pelvic findings before and after the injection somewhat equivocal.

OBSERVATIONS

We have made observations in 119 cases to determine the reaction of the uterus to intravenous pitocin. If no definite softening of the uterus was evident before the injection, any change in consistency which may have occurred after the injection was so slight that it could not be recognized by palpation. For this reason we were unable to observe a definite change in the consistency of the normal non-pregnant uterus after the injection of pitocin. Moir (12) states that the oxytocic principle of pituitrin in a dose of 10 units appears to have no effect on the non-pregnant uterus. That the non-pregnant uterus is stimulated by pitocin to contract is demonstrated clinically by the fact that severe metrorrhagia may be alleviated by this oxytocic when administered intranasally or hypodermically. However, where there was definite softening of the uterus due to pregnancy, the uterus became almost ligneous in consistency within one minute after the injection. When fibroids were present in the pregnant uterus it was observed that these tumors were outlined with difficulty after the injec-

tion of pitocin due to the increase in the consistency of the softened portion of the uterus. After the injection of pitocin in one patient with retroversion, the uterus was found in an anterior position as a result of the increase in the consistency of the uterus and the contraction of the round ligaments. The change in the consistency of the uterus is especially striking at the level of the internal os, as a result of which Hegar's sign disappears and the lower part of the uterus and the cervix become an easily palpable finger-like structure. In this connection reference should be made to a statement by Geiling (13) that "during the early stages of pregnancy the human uterus does not react to pitocin, probably because of the inhibitory effect of the luteal secretion." In our experience there is an increase in the consistency of the pregnant uterus after the injection of pitocin which is obviously due to the contraction of the muscle. This does not necessarily contradict the inhibitory effect of the corpus luteum inasmuch as the dosage of the 0.5 cc. of pitocin may not be physiologic and is sufficient to counteract the inhibitory effect of the corpus luteum as described by Knaus, Robson (14), Reynolds (15) and others.

In the group of 119 patients there were 17 in whom a menstrual period had not been missed at the time of the observation. In 9 of these women there was an increase in the consistency of the uterus after the injection and a diagnosis of pregnancy was made which was confirmed in only 4 instances by the subsequent histories. In the 5 women who were not pregnant the softening of the uterus was undoubtedly due to the premenstrual phase. In 8 cases no change in the consistency of the uterus was noted. Of these, 2 were later found to be pregnant. In 7 cases of this group the response of the uterus to the oxytocic agent was either of no value in detecting the presence of a pregnancy or led to a diagnosis of pregnancy where none existed. This amounts to an error of 41 per cent.

In the next group there were 5 patients in whom a menstrual period had been missed with an interval of 30 days or less since the onset of the preceding period. In 3 instances there was a marked increase in the consistency of the uterus following the intravenous injection of pitocin and in these 3 patients pregnancy was subsequently shown to have been present. In 1 case there was no change in the consistency of the uterus and the following course showed that pregnancy had not been present. In the case of the other patient who was pregnant, no change in the consistency of the uterus was noted after the injection of pitocin. This patient was difficult to examine satisfactorily because of obesity and marked pelvic tenderness resulting from a ruptured ectopic pregnancy. In this group of 5 cases there was 1 error, or an error of 20 per cent.

In another group there were 56 women in whom 31 to 60 days had elapsed since the onset of the last menstruation. In 5 instances there was a definite increase in the consistency of the uterus after the injection, although no pregnancy was present. In 44 cases there was an increase in consistency of the uterus after the injection. However, in these patients the soft consistency of the uterus which was found on the first examination naturally suggested the possibility of a pregnancy. In 4 other cases no definite softening of the uterus was noted on the first examination and no increase in consistency was noted

after the injection, although these 4 patients were later shown to have been pregnant. In 3 cases no change in the consistency of the uterus was noted after the injection and the subsequent course in each demonstrated that there had been no pregnancy. In this group there was an error in 9 cases or 16 per cent.

In 29 patients in whom 61 to 90 days had elapsed since the onset of the last menstruation, there was an increase in the consistency of the uterus after the injection in 25 instances. In these 25 patients the pelvic findings prior to the injection indicated the probability of pregnancy and this was confirmed in each case by the subsequent course. In another patient 46 years of age, a subinvoluted uterus was thought to be slightly softened prior to the injection, after which there was a definite increase in consistency. In 2 cases of pregnancy no change in consistency was noted after the injection. In another patient who was not pregnant no change in the consistency of the uterus was elicited after the injection of the oxytocic agent. In this group there was an error of 10 per cent.

There were 12 patients in whom 90 days or more had elapsed since the onset of the last period. In 9 of these in whom a tentative diagnosis of pregnancy was made from the pelvic findings, there occurred an increase in consistency following the injection. Pregnancy was later demonstrated in these patients. In the remaining 3 patients there was no softening of the uterus on the first examination and no change in consistency was noted after the injection. These patients were not pregnant. There was no error in this group.

In 2 cases a large abdominal tumor was found on palpation. There were no other signs of pregnancy in either case, although 1 patient had not menstruated for 8 months. After the injection of pitocin no change in the consistency of the tumor was noted. In each instance a large degenerating fibroid was demonstrated at operation. Others have observed that a soft, degenerating fibroid does not show any change in consistency after the injection of an oxytocic agent. This negative evidence is of considerable help in eliminating the possibility of pregnancy.

In this series of 119 patients of whom 74 were pregnant, there were 4 abortions following the injection of pitocin. One patient aborted a 4 months fetus 5 weeks after the injection of pitocin. Another patient who was 2 months pregnant, aborted the day after the injection. Another patient who was 3 months pregnant aborted 7 days after the injection. A fourth patient who passed a fetus of 4 months' gestation had received the injection of pitocin 64 days prior to the abortion. The incidence of abortions is 5.3 per cent. Since this is a relatively low incidence of abortion, it does not seem probable that the intravenous injection of pitocin was directly responsible for the abortions in this group of patients. This is also in agreement with the experience of other observers.

There were 4 patients who gave a history suggesting ectopic pregnancy. In 1 there was found to be an increase in the consistency of the uterus after the injection, although the subsequent course of the case demonstrated that no ectopic pregnancy was present. In a similar case no contraction of the uterus was noted after the injection and the follow-up in this case demonstrated

that no ectopic pregnancy was present. In 2 other cases where an ectopic pregnancy was present, an increase in the consistency of the uterus was noted in only 1 case. In this group of 4 cases of ectopic pregnancy and suspected ectopic pregnancy, the reaction of the uterus to pitocin was helpful in making the differential diagnosis in only 2 cases.

In 1 case of retroversionflexion the uterus came forward spontaneously after the injection. This has been described by Reeb who suggests the intravenous injections of an oxytocic for the purpose of replacing the retroflexed gravid uterus. In another case of retroflexion manual replacement of the uterus was accomplished only after the injection. In another patient with a retroflexion the pregnant uterus was much more easily palpated after the injection which made it possible to definitely outline the size and shape of the uterus. This has usually been found to be the case, irrespective of the size or position of the uterus. In view of the increased ease with which the uterus can be palpated after the injection, the application of this method would be a distinct aid to students who are inexperienced in making pelvic examinations.

After the injection of pitocin there were subjective symptoms in 41 cases. The commonest complaint was headache. Others complained of a feeling of warmth, palpitation and a feeling of fullness in the head. These symptoms usually subsided within 5 minutes. In no case was there more than a slight rise in blood pressure following the injection.

CONCLUSIONS

The response of the uterus in early pregnancy to the intravenous injection of pitocin is an increase in consistency. Following the intravenous injection of pitocin the normal non-pregnant uterus shows no definite change in consistency which can be elicited by palpation.

In all the cases in which there was a marked change in the consistency of the uterus following the injection of pitocin the previous pelvic examination invariably showed softening of the uterus which is a probable sign of pregnancy.

In cases of ectopic pregnancy there was difficulty in demonstrating softening of the uterus.

The intravenous injection of pitocin carries no appreciable danger for the pregnancy or the patient. One-third of the patients complained of headache and a transient feeling of apprehension following the injection of pitocin intravenously.

Degenerating fibroids do not change in consistency after the intravenous injection of pitocin.

The reaction of the pregnant uterus to pitocin is a helpful aid in the differential diagnosis of pregnancy but is not a conclusive test for pregnancy.

The reaction of the premenstrual uterus to pitocin needs further investigation.

SUMMARY

In the series of cases reported there was no evidence that the intravenous injection of pitocin involved any risk to the pregnancy. Within one minute

after the injection the softened pregnant uterus shows on palpation an increase in consistency of such degree that it may be described as ligneous. In a few cases this reaction has been observed where a menstrual period had not as yet been missed. In our experience degenerating fibroids do not show any change in consistency after the injection, which is helpful in making the differential diagnosis.

REFERENCES

1. Hoehne-Greifswald: Arch. f. Gynäk. **125:** 356. 1925.
2. Lorincz, B.: München. med. Wchnschr. **38:** 1621. 1927.
3. Zorn, W.: Zentralbl. f. Gynäk. **20:** 1315. 1926.
4. Rodecurt, M.: Deutsche med. Wchnschr. **57:** 976. 1931.
5. Reeb, M.: Gynéc. et obst. **21:** 193. 1930.
6. Kustner, H.: Deutsche med. Wchnschr. **56:** 384. 1930.
7. Knaus, H.: Arch. f. exper. Path. u. Pharmakol. **124:** 152. 1927.
8. Knaus, H.: Zentralbl. f. Gynäk. **53:** 2193. 1929.
9. Reynolds, S. M. R.: Am. J. Physiol. **97:** 706. 1931.
10. Reynolds, S. M. R., and M. H. Friedman: Am. J. of Physiol. **94:** 696. 1930.
11. Fellner, O. O.: Zentralbl. f. Gynäk. **49:** 869. 1925.
12. Moir, C.: Edinburgh M. J. **49:** 93. 1934.
13. Geiling, E. M. K.: J. A. M. A. **104:** 738. 1935.
14. Robson, M.: J. Obst. & Gynaec. Brit. Emp. **49:** 498. 1933.
15. Reynolds, S. M. R.: Am. J. Obst. & Gynec. **29:** 630. 1935.

STUDIES ON OVARIAN DYSFUNCTION. III. THE MENOPAUSE[1]

FULLER ALBRIGHT
Massachusetts General Hospital
BOSTON, MASSACHUSETTS

Science rightly concluded many years ago that the phenomenon called the menopause is associated with a cessation of ovarian function. The structure of the ovary after the menopause, the atrophy of the uterus and vagina during the menopause, the close analogy between physiological menopause and artificial menopause were all evidence of this. But ovarian function, it is now known, may cease because the ovary per se ceases to function or because the stimulating hormone or hormones from the anterior pituitary cease to be furnished. With which of these possibilities is one confronted at the menopause?

Evidence that ovary is primarily and not secondarily hypofunctioning at the menopause. In the previous paper of this series (1) it was pointed out how the determination of certain hormones in the urine could be used in differentiating between a hypogonadism of primary ovarian cause and one secondary to a hypofunction of the gonad-stimulating hormone or hormones of the anterior pituitary. Obviously in both instances there would be a cessation of menses, and atrophy of ovaries, uterus, and vagina. The differential point is that, if the ovary is primarily at fault, the gonad-stimulating hormone of the anterior pituitary not only continues to be formed, but, as Zondek first showed (2), is excreted in increased amounts.

In paper I (3) of this series the results of the prolan A test on normal individuals and the methods used in our laboratory were recorded. It was

TABLE 1. RESULTS OF TESTS IN THREE STATES UNDER CONSIDERATION

	"E" Test	"PA" Test
Normal State	+	0
Hypogonadism—primary hypoovarian	0	+
Hypogonadism—secondary to pituitary hypofunction	0	0

there pointed out that each laboratory must have its own standard of normals due to the many possibilities of varying the technique. Therefore, hereafter the designations "PA" and "E" for prolan A and estrin tests respectively as done in this laboratory will be used. Briefly stated it was found that in normal individuals the E test made at random anywhere during the cycle is positive about four times out of five, and the PA test is negative with very rare exceptions.

Table 1 shows how these tests can be used in differentiating hypogonadism from the normal state and in distinguishing the two types of hypogonadism.

[1] From the Ovarian Dysfunction Clinic of the Massachusetts General Hospital.
Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 11, 1935.
All estrin therapy used in these studies was given in the form of Progynon pills, kindly supplied by the Schering Corporation of Bloomfield, New Jersey.
This investigation was made possible by grants from the DeLamar Mobile Research Fund and the Proctor Fund of the Harvard Medical School.

These tests applied to patients at the menopause leave no doubt that the hypogonadism is due to the ovary ceasing to respond to the pituitary hormone. Patients with spontaneous menopause, like gonadectomized patients, have positive PA tests and negative E tests. Our experience here is merely confirmative of that in the literature (2), (4). Thus the fact that the hormonal pattern at the menopause is the same as that following bilateral oöphorectomy is further evidence that the ovaries are primarily at fault—that the menopause is a physiological "ovarian amenorrhoea".

Are vasomotor symptoms directly due to hypoestrinism, or excess of prolan A or a third factor? The next question has to do with the symptoms of the menopause. We will limit the discussion to the symptoms at the "hot flash stage", and by symptoms will mean the vaso-motor phenomena of which the hot flash is the most striking. At this stage of the menopause a patient has symptoms and at least two changes as regards her hormone pattern: (a) hypoestrinism; (b) an overproduction of prolan A. A priori the symptoms might be due to either (a) or (b) or to a third variable. The present study was arranged to determine this question. The plan of the study was simply to make frequent E tests and PA tests before, during, and after treatment with estrin and to plot the number of hot flashes against the results of the tests. In this way we hoped to be able to determine whether the flashes are proportional to the positivity of the PA tests or the negativity of the E tests, or independent of either.

DISCUSSION OF DATA

Observations have been made on over thirty individuals. The data on ten of these are presented in detail. Certain conclusions can be made; certain speculations strongly suggest themselves. In order that the reader may understand the basis for the conclusions and speculations suggested, the chart in figure 1 has been constructed. It represents how the three variables under consideration would probably fluctuate in a normal woman who first was subjected to gonadectomy and who then, after a prolonged interval, received large doses of estrin. This chart is diagrammatic, but is constructed on the basis of the data to be presented later. It is introduced ahead of the data in order that the reader may appreciate the significance of the individual observations as they are presented.

In figure 1 the three variables may be examined one at a time. The estrin excretion in the urine is at a certain level before gonadectomy and there is a positive E test; after gonadectomy estrin immediately disappears from the urine and the E test becomes negative; with sufficient estrin treatment, estrin immediately appears in the urine and the E tests become even more strongly positive than in normal individuals. The prolan A in the urine before operation is very slight, giving a negative PA test; it rises gradually after operation eventually getting high enough to give a positive PA test and finally reaching a plateau where it remains indefinitely unless treatment is instituted; with estrin treatment the prolan A in the urine decreases and eventually gets below the threshhold for a positive PA test. The hot flashes do not appear until after a definite latent period following gonadectomy; they then rise to a maxi-

mum following which they gradually decline so that they would disappear after a period of months or years if no treatment were started; with treatment there is a definite latent period again before the hot flashes diminish. The shapes of the curves in figure 1 are entirely speculative as the data are not sufficiently accurate for one to plot smooth curves.

The question can now be examined as to whether the hot flashes can be correlated with the lack of estrin excretion or the increase in prolan A excretion or neither.

As regards lack of estrin, there are two obvious reasons why this is not

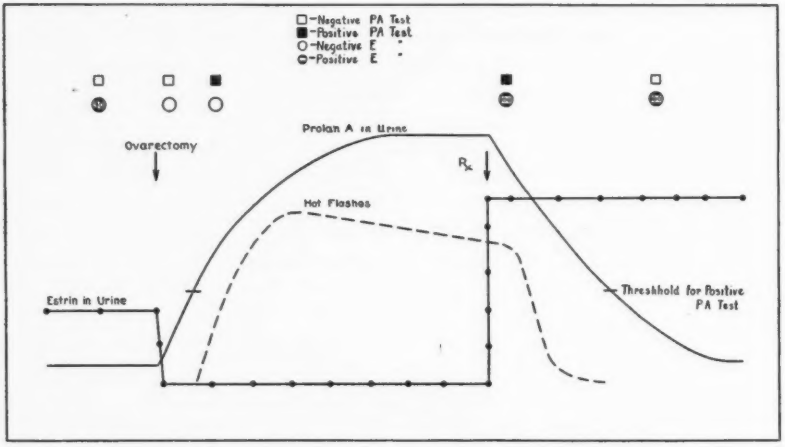


Fig. 1. Schematic chart representing the main conclusions which the data suggest (see text). Chart depicts how the three variables under consideration—estrin excretion in urine, prolan A excretion in urine, and number of hot flashes—vary with one another during the normal state, following castration, and following subsequent estrin replacement therapy.

the direct cause of the hot flashes. Estrin disappears in many cases following gonadectomy long before the hot flashes begin and, conversely, the system can be flooded with estrin as the result of therapy long before the hot flashes disappear (see fig. 1). As pointed out in paper II of this series (1), cessation of periods as a result of pituitary hypofunction is not accompanied by hot flashes. Here there is the same lack of estrin but a lack rather than an increase in prolan A. The tendency for the symptoms to come in outbursts is more consistent with periodic discharge of a substance produced in excess rather than to the complete absence of a substance. Our studies leave no question, however, concerning the fact that estrin therapy in sufficient doses and over a long enough time does make the hot flashes disappear.

When one tries to correlate the hot flashes with the positivity of the PA tests, one has a little better success. The two curves do rise and fall at the same time. One notes, however, that the prolan A curve reaches a plateau while the hot flash curve slopes downward. Therefore, if the hot flashes are directly due to the prolan A, one would have to make the further not unlikely assumption that the vasomotor system adjusts itself in time to a given

quantity of prolan A. This would explain why the same amount of prolan A might produce more hot flashes shortly after gonadectomy than later on after treatment had been instituted (see fig. 1). One reason for believing the hot flashes are directly due to the prolan A is that patients whose hot flashes are very easily affected by treatment have PA tests which fluctuate very easily with treatment. The opposite is true in patients who are very resistent to treatment (cf. exhibit IV and exhibit VI). That the hot flashes are directly due to the excessive prolan, though not unlikely, remains unproven. The possibility remains that some other factor is directly responsible for the symptoms and is modified by estrin in a manner almost parallel to prolan A.

Before presenting the actual data two criticisms of this work should be made. In the first place, the term estrin is used as if it referred to a specific substance. It is now known that there are at least three estrogenic substances occurring in women. It is just possible that lack of one of these might be the cause of the hot flashes, and that this might not be apparent where the three are treated indiscriminately. Secondly, as will appear in the data, the PA tests every now and then are negative when one would have expected a positive test and vice versa. The preponderance of tests agree with the argument here presented. It is hoped that the points of disagreement are due to the admittedly rough technique of the test and not to a fault in the argument.*

DATA

The data are presented by means of 11 exhibits.

Exhibit I. Showing that administration of estrin by mouth in doses used for treatment is immediately followed by excretion of estrin greater than normal. The patient was 55 years old. The periods became irregular three years previously and stopped one year previously. The hot flashes were moderate in amount.

Date	E test	PA test	Hot flashes	Estrin B
6-17-34	0	neg.	4	—
6-18-34	0	"	4	—
6-19-34	30+U.**	strongly pos.	16	600 U. t.i.d.
6-20-34	44+U.	pos.	7	" " "
6-21-34	26+U.	weakly pos.	2	" " "
6-22-34	34+U.	" "	9	" " "
6-23-34	46+U.	0*	10	600 U. i.d.
6-24-34	15+U.	+	4	" " "
6-25-34	35+U.	0	Not counted	" " "
6-26-34	35+U.	+	" "	" " "

* In some of the later experiments the PA test was made with four different dilutions, each dilution being twice as weak as the preceding one. These roughly quantitative PA tests will be reported as 0, +, ++, +++, ++++, instead of as positive or negative. In terms of mouse units, these correspond to 0, 8, 16, 32, and 64 units per 100 cc. of urine.

** The "+" sign indicates tests were positive for greatest dilution used but that tests were not carried out on further dilutions.

These data show that on the first day of estrin administration this hormone was excreted in the urine in amounts as great as, and probably greater than, normal,

* When these data were collected the importance of the pH in the precipitation of prolan A from the urine by alcohol was not appreciated. Since the alcohol used in the laboratory for this purpose had a slightly alkaline value it is probable that the tests here reported were carried out at a more optimal pH value than otherwise would have been the case.

showing that it is possible with such doses immediately to flood the system. Hot flashes did not immediately disappear, however. This particular patient had an unusually weak PA test and very few vasomotor phenomena.

Exhibit II. Spontaneous menopause in woman of 52 years. This patient when first seen had had no periods for about one year and hot flashes for about one and one-half years. The studies on her are shown in figure 2.

One notes the relatively long time required for the disappearance of the hot flashes and also for the appearance of negative PA tests. The presence of hot flashes during treatment, with E tests more strongly positive than in normal women, sup-

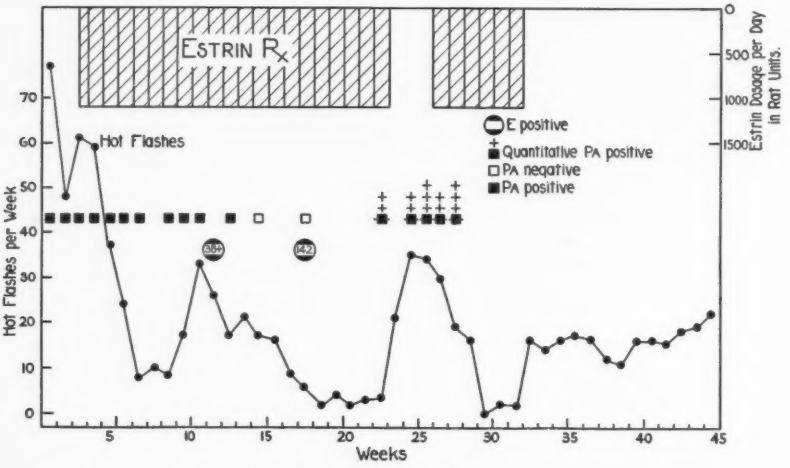


Fig. 2. Data for Exhibit II.

ports the contention that lack of estrin is not the immediate cause of the hot flashes. The positive prolan test in the 23rd week is an example of the fact that this test does not always parallel the hot flashes.

Exhibit III. Artificial menopause in a patient of 30 years. In November 1932 "both ovaries were removed", without the uterus, at another hospital because of the probably erroneous diagnosis of chocolate cysts. Hot flashes began in December 1932. From January to June 1933 she had regular periods every month, showing that some ovarian tissue must have been retained. Periods then stopped and never recurred. When first seen in January 1934 she was feeling very tired and was having one to five hot flashes daily. The studies on this patient are shown in figure 3.

Since it was known that this patient still had some ovarian tissue left, in spite of one negative estrin test (q.v.), it seemed wise to try to stimulate this before starting on estrin treatment. She received 100 units of Antophysin[1] daily for five days during period 3. The estrin test at the end of this time was still negative so it was concluded that this treatment was without avail.

This study was included, however, chiefly to bring out a point in estrin dosage. During periods seven to ten, 600 units of estrin daily did not decrease the number

[1] Antophysin, supplied by the Winthrop Chemical Company, has been used in our clinic as a source of A.P.L. hormone.

of hot flashes significantly. With 1800 units a day the hot flashes were quickly reduced to none at all. They were then held in check with smaller and smaller doses, finally only 200 units a day being used. The prolan tests decreased in an unusually orderly fashion, showing that time as well as dosage is important in obtaining negative tests. The hot flashes did return later on when the estrin treatment was entirely omitted.

Exhibit IV. Artificial menopause in woman of 27. Both ovaries were removed on September 17, 1934. Hot flashes began in about two weeks. The patient was first seen on November 13, 1934 when the studies were started (see fig. 4).

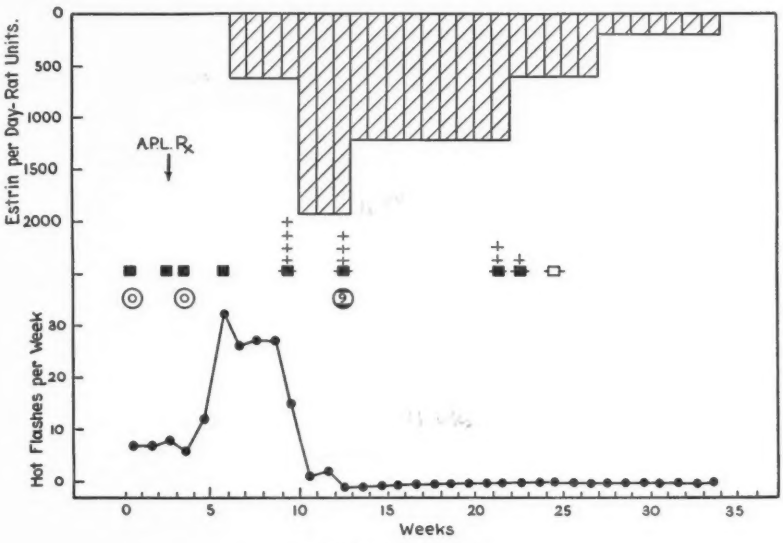


Fig. 3. Data for Exhibit III.

This study differs from most of the others in that the hot flashes fluctuate much more rapidly than usual with administration or withdrawal of estrin therapy. Because of this the hot flashes are charted in three-day periods instead of seven-day periods. Corresponding with this lability of the hot flashes one notes a lability of the prolan excretion, which is fair circumstantial evidence that the two are related.

The fact that the prolan tests were negative in spite of the presence of hot flashes when the patient was first seen (periods 20-32) does not necessarily mean that the hot flashes are not due to the prolan, as the latter may be above normal and still below the threshhold for a positive test (see above).

The rapidity with which estrin becomes positive and negative in the urine following administration or cessation of treatment respectively is well shown (e.g. periods 29 and 32). Certain discrepancies in the prolan test do appear (cf. negative test in period 61). Note that the patient had a period following estrin treatment (periods 77 and 78).

During periods 84, 85, 86, 88, 89 a different preparation of estrin was used, which turned out to be entirely ineffective as regards alleviation of symptoms or ability to decrease prolan A excretion or produce estrin in the urine. There have been too few observations made with this preparation, however, to justify the pub-

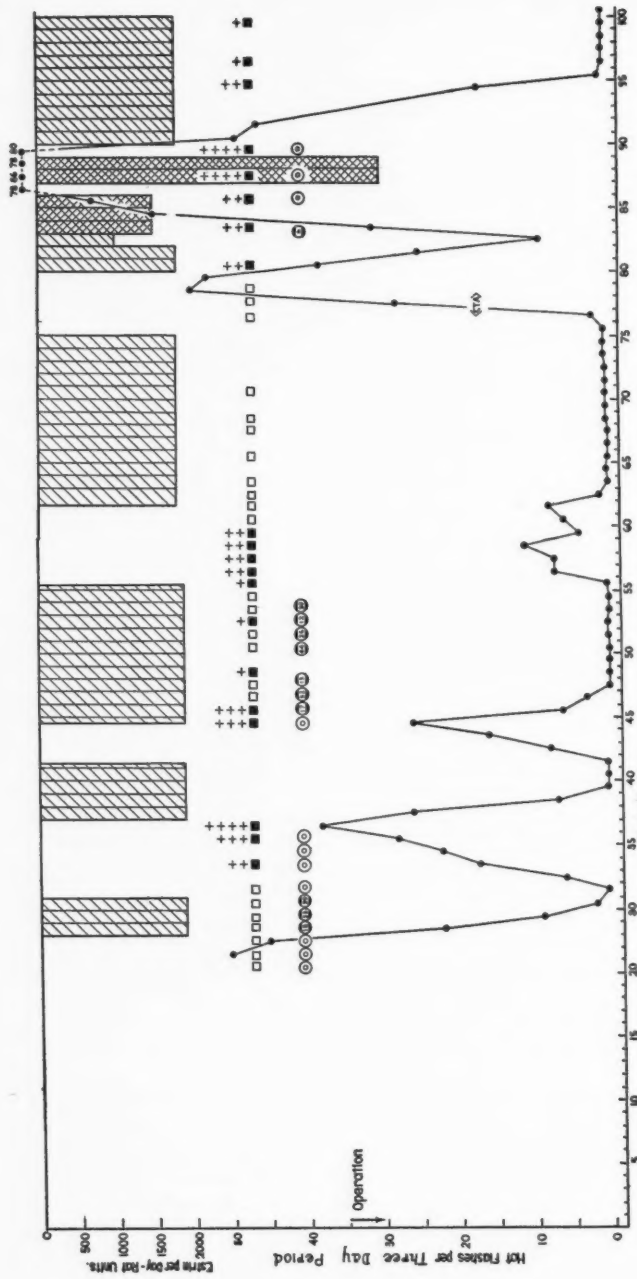


Fig. 4. Data for Exhibit IV.

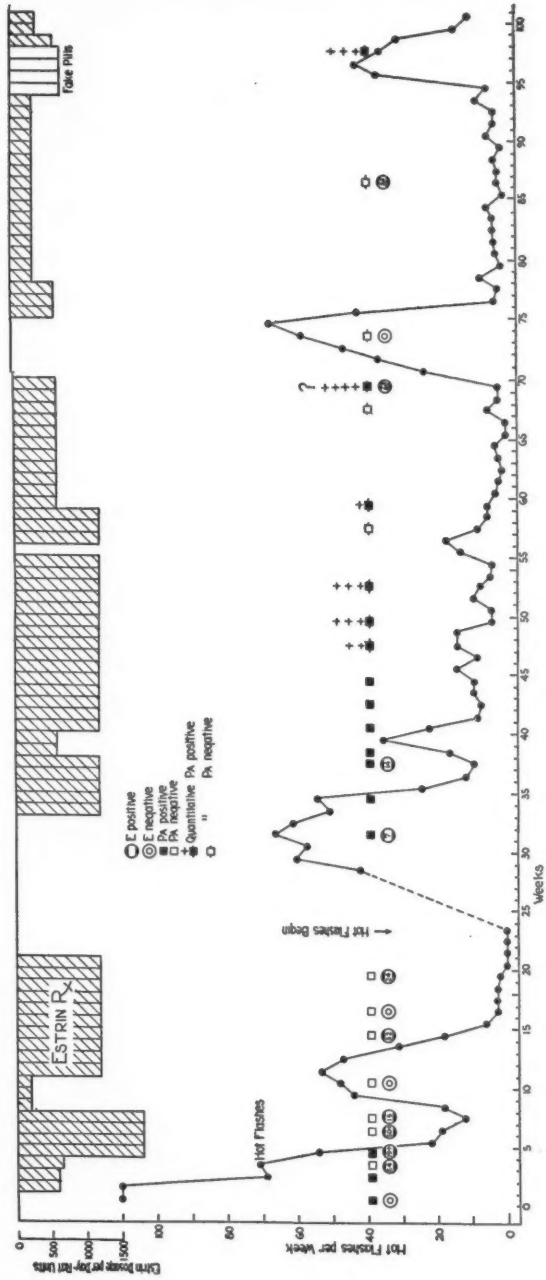


Fig. 5. Data for Exhibit V.

lication of its name. It might be that something happened to this particular allotment of the drug, rendering it ineffective. In the chart this preparation is indicated by cross-hatched columns.

Exhibit V. Spontaneous menopause in patient of 52 years. This patient's menopause began in 1931. She was first seen on May 29, 1933 when the studies were started which have been continued for two years (see fig. 5).

This study is of particular interest as regards the duration of treatment necessary and the dosage. One notes that the hot flashes always recur when treatment is discontinued, although the dosage to prevent recurrence becomes less and less. Thus, during periods 39 and 40, 600 units of estrin a day was not sufficient to prevent an

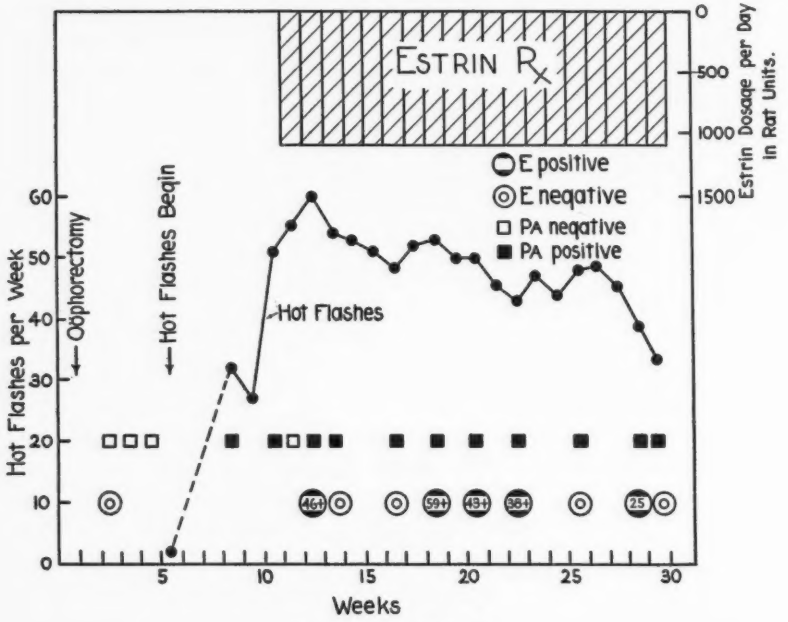


Fig. 6. Data for Exhibit VI.

increase in the number of hot flashes; during periods 59-70 the same dose was sufficient; during periods 76-95, 400 units were sufficient. The strongly positive PA test during period 69 may have been incorrect as there is reason other than that it does not fit the argument to question its accuracy. In order to rule out any psychological effect which the pills might be having Dr. Stragnell of the Schering Corporation was kind enough to have some pills made up with the same exterior but containing no hormone. These "fake pills" did not stop the hot flashes. The fact that the patient still had hot flashes with negative prolan tests during period 11 and had only a few with strongly positive prolan tests during periods 43-53 is against the hypothesis that the hot flashes are due to the prolan A.

Exhibit VI. Artificial menopause in patient of 31 years. This patient had her remaining ovary removed for dermoid cyst on July 19, 1933 at which time the studies were started (see fig. 6).

This study is of interest in that the hot flashes were peculiarly resistent to treatment and the prolan test never became negative. This is the exact opposite of the situation seen in exhibit IV (q.v.). One notes in addition that the hot flashes appeared about the time that the prolan tests became positive. Why four of the estrin tests should have been negative during treatment while the remaining ones were strongly positive is difficult to understand.

Exhibit VII. Spontaneous menopause in patient of 47 years. When first seen on February 27, 1933 this patient had been having hot flashes for six years. In addition, she had had her uterus and all but a small piece of one ovary removed two years previously. The operation had not aggravated her menopausal symptoms. Her studies from July 1933 on are shown in figure 7.

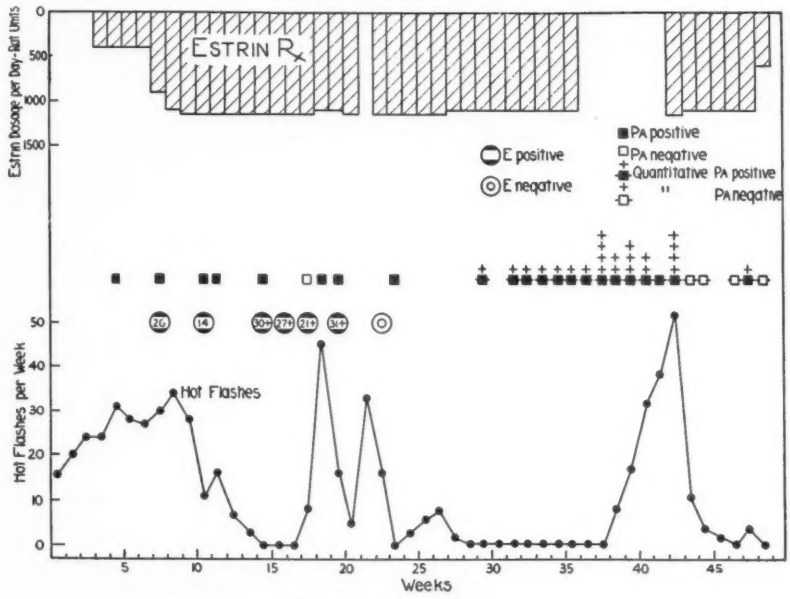


Fig. 7. Data for Exhibit VII.

The prolan and estrin tests agree very well on the whole with the principles suggested in the schematic chart. Why there was a sudden increase in hot flashes during periods 18 and 19 is hard to say. The increase during period 22 was apparently due to cessation of therapy. The suddenness of this rise is unusual. The step-like rise over a period of seven weeks during periods 37-43 is more characteristic of the rise following cessation of treatment. The rapidity of the fall in the number of hot flashes with treatment during periods 43-45 is in agreement with the rapidity with which the prolan tests became negative.

Exhibit VIII. Patient of 32 years who received x-ray sterilization. This patient had an operation for cancer of the breast on September 19, 1932. She received x-ray sterilization during October 1932 in the belief that cessation of estrin production would decrease the chance of recurrence. Her hot flashes started in November 1932

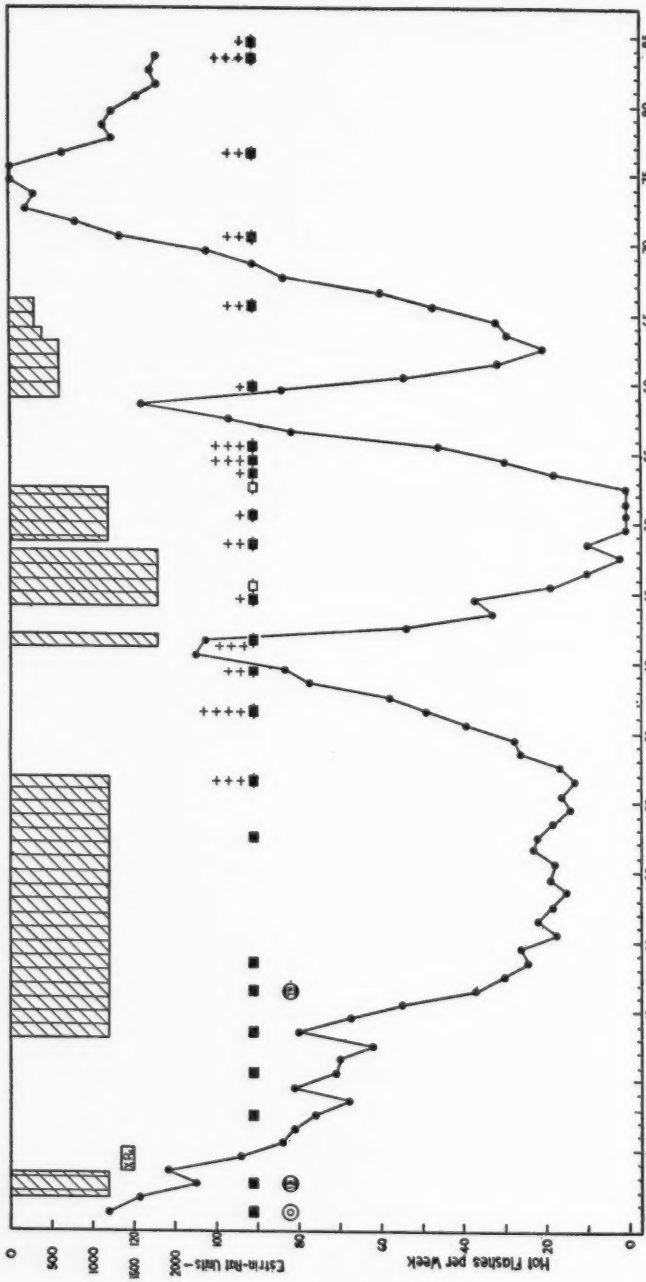


Fig. 8. Data for Exhibit VIII.

and she was first seen during July 1933 when studies were commenced (see fig. 8).

Prolonged estrin treatment was contraindicated in this patient because of the possible effect of estrin on breast cancer. Since the patient was extremely miserable, it was decided to try irradiation of the pituitary to see whether this would decrease the prolan A overproduction and perhaps the hot flashes. She received treatment during periods 4 and 5. Her hot flashes were somewhat reduced (cf. periods 4-14), but not convincingly so and the prolan test never became negative. Unfortunately no attempt to quantitate these tests was made. No conclusions can be drawn from this experiment. As far as it goes it favors the theory that hot flashes are due to prolan A. It also demonstrates that x-raying the pituitary is hardly worth while from a therapeutic point of view.

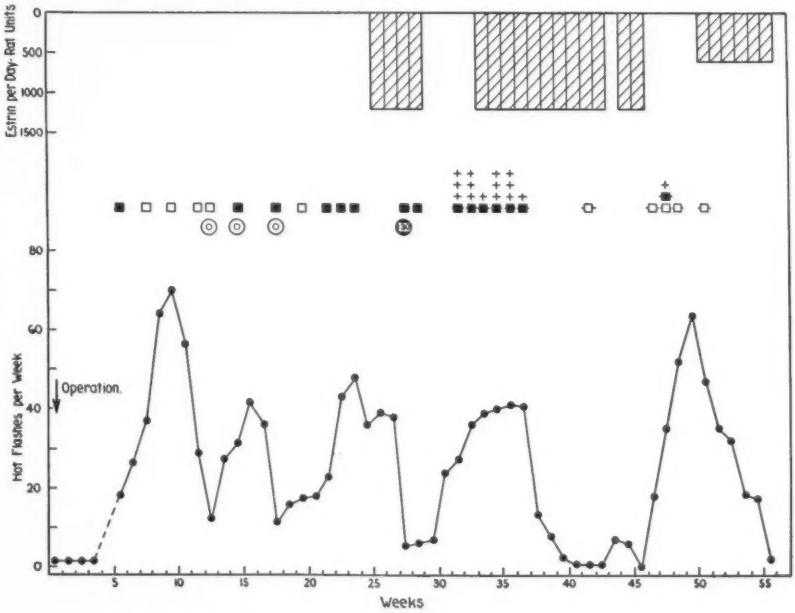


Fig. 11. Data for Exhibit XI.

Periods 67-87 are of interest as a control to see what happens without treatment. This patient shows perhaps better than any other one the orderliness of the increase and decrease in the number of the hot flashes, week by week. Thus it took ten weeks for the flashes to reach their peak after medicine was stopped the last time (see periods 63-72). This period of time is not consistent with estrin disappearing from the body, but is consistent with the time it might take prolan A production to reach its height in response to lack of estrin.

Exhibit IX. Artificial menopause in patient of 33 years. This patient had a bilateral oophorectomy on August 12, 1933 aand was first seen on September 18, 1933 just after hot flashes had begun.

This study (see fig. 9) is included particularly because of the long control period without treatment. It is of interest that both the hot flashes and the prolan

tests showed some spontaneous fluctuations. The fluctuations did not coincide particularly well. The four weeks latent period in overcoming the hot flashes during periods 34-37 agrees with the continued high amount of prolan in the urine. The fact that this patient had more hot flashes during period 49 with a negative prolan test than during period 36 with a strongly positive one is against the theory that prolan causes the hot flashes, but again may be explained by the contention that the vasomotor system had adjusted itself to continued high prolan production in the latter instance.

Exhibit X. Spontaneous menopause in a patient of 53 years. This patient was first seen in March 1934, her menopause at that time having continued for five years. She was still having about four hot flashes daily (see fig. 10).

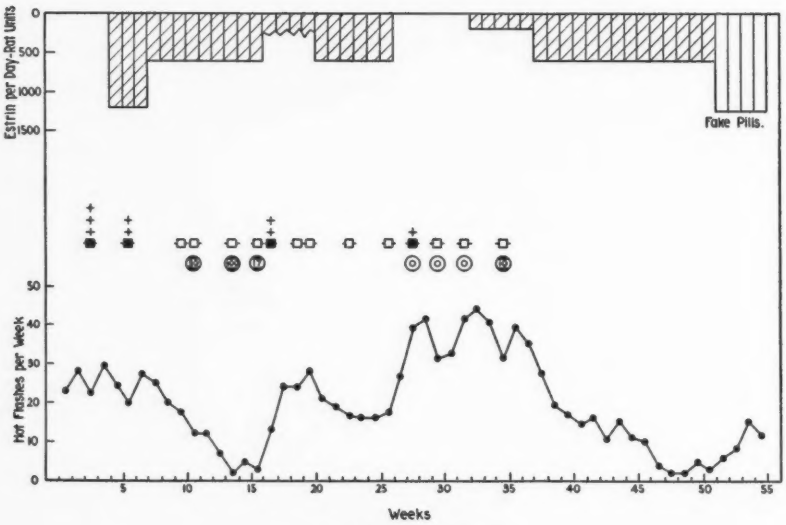


Fig. 10. Data for Exhibit X.

This study is included to bring up an important clinical question. It may be that treating a patient after five years of menopause merely puts her back to where she was five years ago. Thus the prolan tests on this patient soon became negative. When treatment was discontinued, however, the hot flashes immediately rose to higher levels than before treatment, in spite of the fact that the prolan tests were still negative. It may be that a long time spent in adjusting the vasomotor system to high prolan levels or what-not is offset by a short period of treatment. The "fake pills" were without effect. Note that only 200 units of estrin a day caused a positive estrin test in the urine during period 35.

Exhibit XI. Arificial menopause in patient of 39 years. This patient had both ovaries removed in December 1932. Hot flashes started about one month after the operation. Studies were started in March 1934 (see fig. 11).

These data require little comment. One notes that as the hot flashes return dur-

ing periods 16 and 17 the prolan tests remain negative, whereas previously with a positive prolan there had been fewer hot flashes (cf. period 11). This is not an unusual finding. It may be that after a period of decreased prolan production the vasomotor system is hypersensitive so that it reacts to amounts which are greater than normal but below the threshhold for a positive test (see above). The "fake pills" were without effect (cf. exhibits V and X). One prolan test before the onset of treatment was negative and unexplainable. Another during the same week was strongly positive.

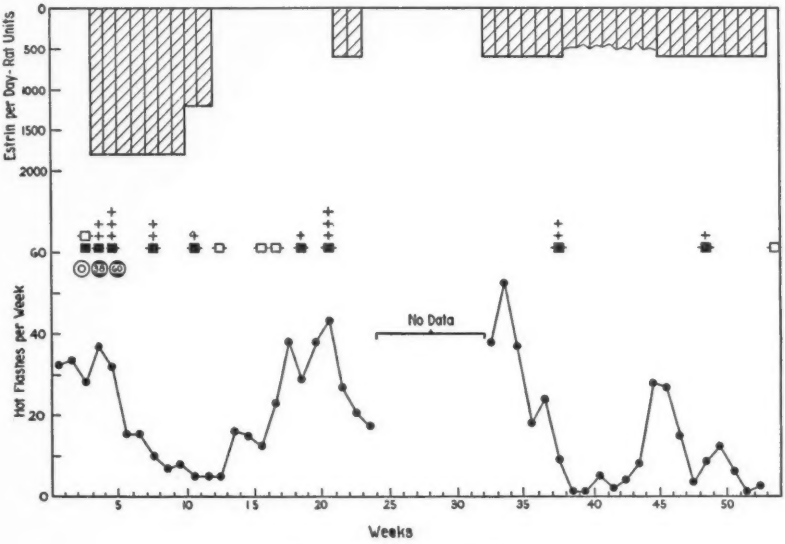


Fig. 11. Data for Exhibit XI.

IMPLICATIONS AS TO TREATMENT

It is not purely an academic question but of therapeutic importance whether estrin exerts its beneficial effect on the symptoms of the menopause directly or through a chain of events as we believe. Whether that "chain of events" is reduction in prolan A production or some other alteration may be immaterial. The very fact that the action is not direct means that length of time of administration may be an important factor. In earlier studies we were almost led to the conclusion that estrin was not effective in the treatment of the menopause because huge doses did not immediately relieve the symptoms. It is now clear that very much smaller doses over a longer period of time will give results. It is also not unlikely that there is a certain dosage of estrin beyond which administration of greater amounts will not speed up the amelioration of symptoms. In other words the result will be dependent on length of administration as well as dosage.

The question of whether one should treat the menopause patient with estrin at all is not too easy to answer. The objection to such treatment would

be that one is just prolonging the period of adjustment. There is probably a real danger in over-treatment. That is, an individual half-adjusted to the hypoovarian condition might be treated with estrin, so that with cessation of treatment she was totally unadjusted again. The sensible thing until further knowledge accrues is probably to give just enough replacement therapy to alleviate but not expel symptoms. In terms of prolan A, if symptoms turn out to be due to this hormone, this would mean to keep the prolan production level low enough so that the patient was not disturbed and gradually allow it to rise as the patient became adjusted to higher and higher levels.

We have not seen any harmful results from long continued administration. One might expect the development of cystic mastitis or metropathia hemorrhagica, but we have not had any such experience. Several patients menstrated a few days after the estrin administration was discontinued. Theoretically estrin is contraindicated in patients who have received x-ray sterilization because of cancer of the uterus, vagina, or breast.

SUMMARY

Data from eleven patients with artificial or spontaneous menopause are reported. The number of hot flashes per unit of time are plotted against the estrin excretions in the urine and the level of prolan A in the urine. The effect of administration and withdrawal of estrin medication on these three variables was noted. The following conclusions are made.

The estimation of the hormones in the urine shows that the hypofunction of the ovaries at the menopause is due to a primary condition in the ovary and is not secondary to hypofunction of the anterior pituitary; that is, the menopause is a physiological "ovarian amenorrhoea."

There are at least two pronounced changes in the hormonal pattern at the menopause; hypoestrinism and overproduction of prolan A. Either of these hormonal abnormalities or a third one might account for the vasomotor symptoms of the menopause.

Hypoestrinism has been shown not to be the direct cause of the vasomotor phenomena, because (a) with oöphorectomy estrin disappears long before the hot flashes begin; (b) during the menopause, the estrin level can be brought with estrin treatment above normal for a long period of time before the hot flashes cease; (c) hypoestrinism due to pituitary disease is not accompanied by hot flashes.

Estrin treatment none the less relieves the vasomotor phenomena of the menopause after a considerable latent period; estrin treatment likewise stops the overproduction of prolan A after a latent period of a similar order of magnitude. Estrin may exert its beneficial effect by decreasing prolan A production, but the evidence that the hot flashes are proportional to the amount of prolan A produced is suggestive rather than conclusive.

Because estrin exerts its beneficial effect at the menopause indirectly, duration of treatment as well as daily dosage is an important factor in the amount of improvement obtained.

REFERENCES

1. Albright, F., and J. A. Halsted: Studies on ovarian dysfunction. II. The application of the "Hormonal Measuring Sticks" to the sorting out and to the treatment of the various types of amenorrhoea. N. England J. Med. **212:** 250. 1935.
2. Zondek, B.: Über die Hormone des Hypophysenvorderlappens. II. Follikelreifungshormon (Prolan A)—Klimakterium-Kastration. Klin. Wchnschr. **9:** 393. 1930.
3. Albright, F., J. A. Halsted, and E. Cloney: Studies on ovarian dysfunction. I. The hormonal "Measuring Sticks" available for clinical use and values obtained on normal individuals. N. England. J. Med. **212:** 192. 1935.
4. Osterreicher, W.: Die Hypophysenvorderlappenhormon bei ausgefallener Keimdrüserfunktion. II. Quantitative Bestimmungen von Sexualhormonen (Hypophysenvorderlappen und Follikelhormone) bei Gesunden, Geistes—und Nervenkranken. Klin.. Wchnschr. **12:** 896. 1933.

EXPERIMENTAL PRODUCTION OF OVARIAN REFRACTORINESS TO ANTERIOR HYPOPHYSEAL STIMULATION IN THE MONKEY*

F. L. HISAW, ROY HERTZ, AND H. L. FEVOLD
Zoology Department, University of Wisconsin
MADISON, WISCONSIN

It has been repeatedly demonstrated that the subcutaneous administration of anterior hypophyseal substance in the female Macacus rhesus monkey brings about marked follicular development in the ovary without accompanying luteinization (1), (2), (3), (4). These studies provide an adequate account of the result of treatment with gonadotropic extracts for a short period of time. The present work is concerned with the phenomena to be observed as a result of prolonged treatment of mature and juvenile female monkeys with unfractionated extracts of the anterior pituitary of sheep and of horses. The extracts were prepared according to the method described by Fevold, et al. (5). The average dose was the equivalent of 0.5 gm. of dried sheep pituitary tissue administered subcutaneously twice daily.

Observations were made upon eleven monkeys; four of these were sexually mature and seven were juvenile. These animals were treated for periods ranging from ten to fifty-one days. A number of animals were allowed to remain untreated for several weeks and were then given a second series of injections. In a few cases a third course of injections were given following a rest period. Daily observations were made with regard to the condition of the external genitalia and sexual skin and daily vaginal smears were taken for microscopic examination. In addition, periodic laparotomies were performed in order to ascertain the size and condition of the ovaries and uterus.

We shall describe separately the phenomena observed in the juvenile and in the mature monkeys, since the response of these two age groups presented certain strikingly different features.

OBSERVATIONS ON JUVENILE MONKEYS

The first external manifestation of a response to the gonadotropic extract in the case of the juvenile monkeys was a slight swelling and reddening of the sexual skin. This usually occurred on about the fifth day of treatment. The sexual skin reached a maximal degree of turgidity and hyperaemia by the tenth to the fifteenth day of treatment, and subsequently regressed to practically its original condition.

Associated with these changes in the sexual skin there occurred fluctuations in the content of the vaginal smear. From the fifth to about the fifteenth day of treatment there was an increasing abundance of desquamated epithelial cells in the smear. Between the fifteenth and twentieth day increasing numbers of leucocytes appeared in the smear almost to the complete exclusion of the

* Assisted by grants from the National Research Council, Committee on Problems of Sex, and the American Academy of Arts and Sciences, Permanent Science Fund Committee.

cornified epithelial cells. Usually on about the twentieth day macroscopic bleeding commenced and continued for approximately six to seven days. During the remainder of the course of treatment the smear showed numerous leucocytes with a few cornified epithelial cells. Microscopic bleeding of 8 to 10 days duration occasionally occurred 20 to 25 days after the cessation of injections.

Four juvenile animals were given a second series of injections after a rest period of several weeks and two animals were given a third course of treatment. These animals presented no external signs of ovarian response throughout the time of these injections. Only one animal showed microscopic bleeding from the twenty-sixth to the thirty-sixth day after the last injection.

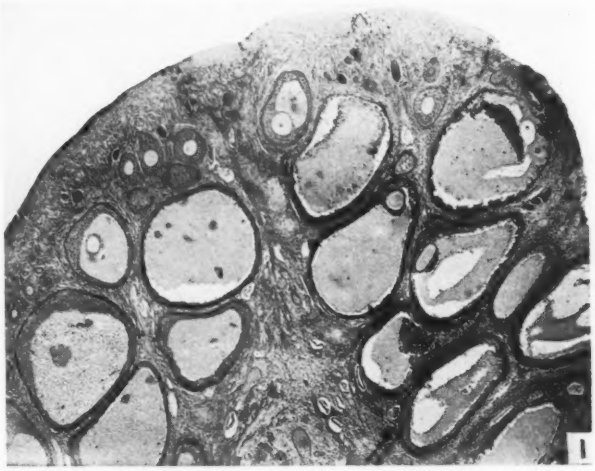


Fig. 1. Juvenile monkey Al.13. Ovary removed 45 days after three series of subcutaneous injections of anterior pituitary extracts. See protocol. The presence of many follicles indicates at least partial recovery from the refractory condition. ×30.

Periodic laparotomies, performed in order to obtain actual ovarian measurements, showed that during the first ten days of treatment the ovaries enlarged usually about five-fold as a result of marked follicular growth. The ovaries then progressively decreased in size and returned to practically their original dimensions by about the twentieth day of treatment. Subsequent administration of the gonadotropic substance led to no ovarian enlargement in the juvenile animal, even in those cases in which a number of weeks had intervened between injections. We have termed such non-responsive ovaries "refractory ovaries".

The following protocol on Monkey Al.13, weighing 2300 grams, presents characteristic results obtained in the juvenile animal:

Experiment 1. Days injected, 36. Total dosage, 35.1 grams equivalent of sheep pituitary powder. Bleeding, 19th to 24th day. Laparotomies, 15th day, Rt. ov. 1.2 × 1.1 × 0.85 cm., Lt. ov. 1.2 × 0.8 × 0.77 cm. 25th day, Rt. ov. 0.95 × 0.6 × 0.5 cm., Lt. ov. 1 × 0.55 × 0.5 cm. 36th day, Rt. ov. 0.8 × 0.6 ×

0.42 cm., Lt. ov. 0.75 × 0.5 × 0.5 cm. Microscopic bleeding 20th to 27th day after discontinuation of treatment.

Experiment 2. Started 41 days after Experiment 1. Days injected, 10. Total

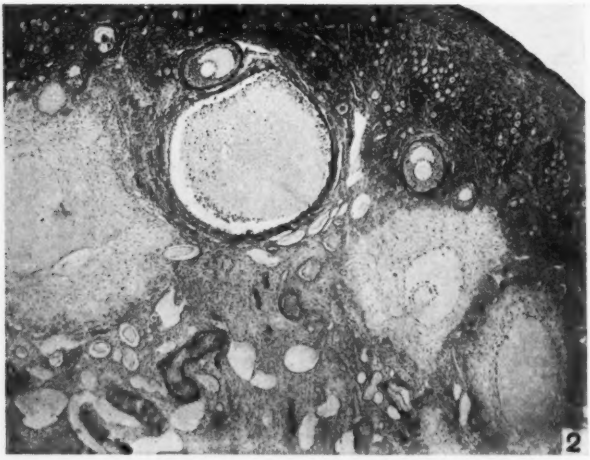


Fig. 2. Sexually mature monkey Al.21. Right ovary. Removed on the 27th day of anterior pituitary treatment. Size .9x.75x.55 cm. Reached maximum size on the 11th day (2.0x1.5x1.5 cm.) Bleeding occurred 16th to 22nd day. The section shows different degrees of follicular degeneration and dilatation of the venous and lymphatic channels. ×30.

Fig. 3. Sexually mature monkey. Al.21. Left ovary. Removed on the 45th day of anterior pituitary treatment. Size .8x.7x.5 cm. Section shows degeneration of all large follicles into cell-poor, hyaline, scar-like bodies. The vascular and lymphatic distension is somewhat reduced. ×30.

dosage, 8 grams equivalent dried sheep pituitary. No change in sexual skin and no bleeding. Ovaries palpated 10th day and found normal in size. Microscopic bleeding 26th to 36th day after discontinuation of treatment.

Experiment 3. Started 139th day after Experiment 2. Days injected, 12. Total dosage, 11.5 grams equivalent dried sheep powder. No modification of sexual

skin and no bleeding. Ovaries palpated 13th day, and found normal in size. No bleeding after treatment. Castrated 45 days later; microscopic bleeding occurred 9th to 11th day after the operation. Ovaries (fig. 1) contained many well developed follicles. Compare with figures 2 and 3.

OBSERVATIONS ON ADULT ANIMALS

In the adult animal the sexual skin attained the maximum degree of turgidity and color within the first 12 to 15 days of treatment. The ovaries similarly reached their maximum size during this time. The subsequent involution of the ovaries was associated with a regression of the sexual skin. Menstrual bleeding of about four or five days duration occurred on about the twenty-seventh day of treatment and in no case did a second bleeding occur even when injections were continued for more than 50 days. The adult animals, in contrast to the juvenile animals, experienced a virtually normal menstrual flow within 30 to 60 days following treatment. Moreover, the adult animals showed ovarian response to subsequent pituitary administration, although the degree of reaction as judged by ovarian size and development of the sexual skin, was never as extreme as during the first series of injections.

The following protocol on Monkey Al.16., weighing 4200 grams, presents results which are typical for the adult animal:

Experiment 1. Started 3 days after normal menstruation. Days injected, 51. Total dosage, 50.5 grams equivalent of sheep pituitary powder. Bleeding 39th to 42nd day. Laparotomies, 20th day, Rt. ov. 1.8 × 1.6 × 1.25 cm., Lt. ov. 1.8 × 1.5 × 1.4 cm.; 32nd day, Rt. ov. 0.95 × 0.9 × 0.65 cm., Lt. ov. 0.9 × 0.85 × 0.7 cm.; 44th day, palpation, ovaries small; 52nd day, Rt. ov. 0.7 × 0.65 × 0.5 cm., Lt. ov. 0.85 × 0.7 × 0.5 cm. Vaginal smears taken for 31 days and no post-treatment bleeding observed. Menstruated 51st to 54th day after treatment.

Experiment 2. Started 59 days after Experiment 1. Days injected, 13. Total dosage, 13 grams equivalent sheep pituitary powder. Bleeding 13th to 16th day. Laparotomy 10th day, Rt. ov. 0.7 × 0.7 × 0.55 cm., Lt. ov. 0.85 × 0.6 × 0.5 cm. No post-treatment bleeding. Menstruated 56th to 60th and 97th to 101st days after treatment.

Experiment 3. Started 110 days after Experiment 2. Days injected, 12. Total dosage, 3.12 grams horse A.P. powder equivalent to 12 grams sheep pituitary powder. No bleeding during treatment and little or no response of sexual skin. Laparotomy 13th day, Rt. ov. 1. × 0.6 × 0.6 cm., Lt. ov. 0.85 × 0.65 × 0.5 cm. Post-treatment bleeding 14th to 16th day. Menstruated 37th to 41st day after treatment.

Experiment 4. Started 140 days after Experiment 3. Days injected 10. Total dosage 10 grams equivalent of sheep pituitary powder. Bleeding 5th to 7th day. Signs of impending menstruation at beginning of experiment. Castrated 11th day. Rt. ov. 0.9 × 0.8 × 0.7 cm., Lt. ov. 1 × 0.9 × 0.7 cm. Post-operative bleeding 21st to 23rd day following operation. No corpora lutea observed during the four experiments.

MICROSCOPIC OBSERVATIONS

Microscopic study of ovaries removed at various stages of these experiments indicates that the reaction of the monkey ovary to continuous hypophyseal stimulation follows a characteristic course. After 10 to 15 days the

ovary is enlarged as much as five-fold. There is extreme vascular engorgement and dilatation of the venous and lymphatic channels. The ovary is constituted primarily of numerous large cystic follicles, practically normal in their histological appearance, except for their excessive distention with follicular fluid. The tunica albuginea is stretched and thinned. The primordial follicles and their contained ova are not markedly affected (6).

After about 20 days the ovary has receded to approximately normal size, but the vascular and lymphatic distention persists. The follicles previously observed are correspondingly reduced in size and are more or less uniform. The granulosa tissue of these follicles presents all degrees of disintegration on down to complete dissolution. In the earlier phases of degeneration the granulosa cells show only hyperchromatic nuclei and a stringy cytoplasm. Later these cells round up and assume the appearance of a lymphoblast, tearing away from the granulosa layer and floating out into the follicular fluid which soon becomes filled with the debris left by these disintegrating cells. Karyorhexis in these cells proceeds quite rapidly and the numerous nuclear fragments often suggest the appearance of a polymorphonuclear leukocyte. However, close scrutiny under high magnification shows the follicles to contain only one type of cell capable of phygocytic activity. These are large round cells of the foam-cell type, possessing a kidney-shaped nucleus and a spongy cytoplasm which appears to contain phagocytized cellular remains. These cells are best seen floating in the cellular debris toward the center of the follicular cavity. Their origin is doubtful, but they are most likely enlarged granulosa cells which have assumed a phagocytic function.

As already stated, in some follicles no trace of the granulosa layers remains. The basement membrane of the granulosa is, however, frequently intact, delimiting the vacated follicular antrum. In those follicles which show fairly complete granulosal degeneration, the thecal layers seem to be next involved. The normal cell-rich connective tissue of the thecae assumes the appearance of embryonic connective tissue, becoming highly oedematous and cell-poor. This swelling advances almost to the extreme border of the follicle, leaving uninvolved only a few of the peripheral fibroblastic bundles of the externa (fig. 2).

As injections are continued for about 45 days these remaining fibroblasts of the externa proceed to lay down increasing amounts of clear-hyaline material which becomes progressively more eosinophilic in character. This hyaline material encroaches upon the oedematous remains of the inner thecal cells, finally replacing the latter, so that the ultimate fate of the large cystic follicles seen in the first stages is their organization into a cell-poor, hyaline, scar-like body which may or may not retain a small trace of the original follicular antrum (fig. 3). It should be emphasized that these changes proceed without any signs of intervening ovulation or luteinization.

Prolonged treatment gives rise to no distinctive vascular changes in the ovary other than the original distention which persists in a somewhat reduced degree throughout the period of injections.

The effect upon the primordial follicles is quite variable. Some cases show

signs of nuclear fragmentation and cytoplasmic granulation in the ova, but this effect is not seen in other cases.

Microscopic examination of uterine tissue obtained by biopsy at the time of bleeding shows that the menstrual flow which occurred during the course of injections was from an endometrium characteristic of the follicular phase of a normal cycle.

DISCUSSION

The menstruation observed during continuous hypophyseal administration is quite closely correlated with ovarian involution and regression of the sexual skin. It is consequently apparent that this bleeding is comparable to the menstrual phenomena previously described by Allen (7) following a reduction in the systemic oestrin level. The oestrus character of the endometrium at the time of the bleeding favors this interpretation.

The negativity of the ovary of the immature animal seems to persist at least for several months, whereas the ovary of the adult monkey seems to partially recover responsiveness after a few weeks of rest. It is quite probable that this difference results from the action of endogenous gonadotropic hormones produced by the adult anterior hypophysis. The practically normal appearing menstruation occurring during the rest period in the adult animal is objective evidence of such hypophyseal activity. The juvenile animal's pituitary seems, however, not to be sufficiently active to effect the necessary restorative processes in the exhausted ovary.

We have previously pointed out (8), (9) the significance of ovarian refractoriness following prolonged hypophyseal stimulation in attempting to account for the clinical occurrences of atrophic gonads in certain instances of hyper-pituitarism such as those described by Cushing (10). We (9) have also shown that gonadal refractoriness may be considered a factor in the progress of senility despite the continued activity of the anterior hypophysis in post-climacteric individuals (Zondek, 11).

The ovarian refractoriness as found in these experiments may have two possible explanations. It may be due to anti-hormone formation as described by Collip (12), Bachman, et al. (13), and Meyer and Gustus (14) or it may be the result of physiological exhaustion of the ovary by over stimulation or a combination of the two. It is highly probable that the refractory condition described here was associated with anti-hormone formation yet the fact that the juvenile monkeys remained refractory after the condition was established while the adults would respond, though to a less degree, after a rest period of two or three months, seems significant. The adults also differed from the juvenile monkeys in that they would recover from the refractory condition and menstruate. If the anti-hormone persisted in the adults it did not prevent the gonad-stimulating action of the animal's own pituitary.

The work of Hertz and Hisaw (9) indicates that the ovaries of young rabbits respond to pituitary extracts in a fashion comparable to that described here for juvenile monkeys, in that the ovaries remain refractory indefinitely after treatment. We have found since that adult rabbits respond like adult

monkeys. That is, their ovaries, within a few weeks, recover sufficiently from the refractory condition to respond again to pituitary treatment. Bachman, et al. (13) found that anti-gonadotropic substances disappeared from the blood of adult rabbits a month or two after discontinuing treatment which is, of course, correlated in time with recovery from the refractory condition. Such recovery, however, in the adult monkey, is not related to the presence or absence of anti-gonadotropic substances. Also, Meyer and Gustus (14) have shown that monkeys whose ovaries have been rendered refractory to pregnant mares' serum will recover and menstruate within several weeks after treatment, while their blood retains anti-mares' serum hormone for at least a year. These observations on rabbits and monkeys seem to indicate that the refractory condition of the ovaries and anti-hormone formation are physiologically independent processes and that anti-gonadotropic hormones formed by injecting sheep pituitary extracts do not inhibit the action of the gonadotropic hormones from the animal's own pituitary.

SUMMARY

Seven juvenile and four adult female Macacus rhesus monkeys were injected subcutaneously with a constant dose of unfractionated anterior pituitary extract for extended periods of time. Several of the animals were allowed extended periods of rest and treated a second and third time. The associated changes in the sexual skin, in the vaginal smear, and in the ovary were followed daily in both the juvenile and the adult animals. A refractory ovarian condition which ultimately obtained persisted in the juvenile animals, but proved only temporary in the adult animals. It is believed that this difference results from the activity of the adult animal's hypophysis in contrast with the inactivity of that of the juvenile animal. The menstruation observed during the course of injections is associated with ovarian involution and external indications of reduced theelin output.

REFERENCES

1. Courrier, R., R. Kehl, and R. Raynaud: Compt. rend. Soc. de biol. **101:** 1093. 1929.
2. Hisaw, F. L., H. L Fevold, and S. L. Leonard: Proc. Soc. Exper. Biol. & Med. **29:** 204. 1931.
3. Saiki, S.: Am. J. Physiol. **100:** 8. 1932.
4. Engle, E. T.: Proc. Soc. Exper. Biol. & Med. **30:** 530. 1933.
5. Fevold, H. L., F. L. Hisaw, A. Hellbaum, and R. Hertz: Am. J. Physiol. **104:** 710. 1933.
6. Hisaw, F. L.: Am. J. Obst. & Gynec. **29:** 638. 1935.
7. Allen, E.: Carnegie Inst. Wash. Pub. No. 380, **19:** 1. 1927.
8. Hisaw, F. L., R. Hertz, A. A. Hellbaum, and H. L. Fevold: Anat. Rec. **55:** (No. 4 Sup.) 1933.
9. Hertz, R., and F. L. Hisaw: Am. J. Physiol. **108:** 1. 1934.
10. Cushing, H.: Bull. Johns Hopkins Hosp. **50:** 137. 1932.
11. Zondek, B.: Die Hormone Des Ovariums; Springer, Berlin, 1931.
12. Collip, J. B.: J. Mt. Sinai Hospital. **1:** 28. 1934.
13. Bachman, A., J. B. Collip, and H. Seyle: Proc. Soc. Exper. Biol. & Med. **32:** 544. 1934.
14. Meyer, R. K., and K. Gustus: Science, **81:** 208. 1935.

THE EFFECTS OF THE TWO OVARIAN HORMONES ON CASTRATED HUMAN FEMALES

C. A. ELDEN

From the Department of Obstetrics and Gynecology, The University of Rochester School of Medicine and Dentistry

ROCHESTER, NEW YORK

Interest in the physiology of menstruation and its associated phenomena in the human female has been given impetus since the isolation and purification of two ovarian hormones. The present communication is concerned with the effects produced by the ovarian hormones on five castrated human females. The observations include: 1) the correlation of the histology of the endometrium produced by estrin alone; and estrin plus progestin; 2) the time relations of bleeding following the use of estrin alone, and of estrin plus progestin; 3) the effect of these hormones on menopausal symptoms.

The estrin (Progynon-B), prepared from mare or stallion urine, was furnished by Schering Corporation. It contained 10,000 R. U. per cc. (50,000 international units) in oil and was used throughout this study. Chemically it is the benzoic acid ester of dihydroestrin.

The progestin was prepared and standardized according to the method of Allen (1), (2) and Allen and Corner (3). Proluton (Schering Corporation) used in this study is an oil solution of progestin, approximately one third as strong as that prepared by the author.

PATIENTS AND EXPERIMENTAL PROCEDURES

The method adopted is that of Kaufmann (4), who succeeded in preparing a premenstrual endometrium and causing an apparently true menstruation in a 17 year old castrated human female. He gave daily intramuscular injections of Progynon-B, for 21 days, a total of 210,000 M. U. (42,000 R. U), followed by 5 Rb. U. (original units) of Proluton for 7 days. In treating primary amenorrheas (5), he gave 10,000 R. U. of Progynon-B twice weekly until 50,000 R. U. had been given, and then followed this with 35-60 Rb. U. of Proluton; beginning 4 days after the last injection of Progynon-B. This latter procedure was first employed here, with control periods of estrin alone. In addition, biopsies were taken at definite intervals.

Case 1. A 24 year old, married female, had a bilateral salpingo-oophorectomy, 9-19-33 for a right ectopic pregnancy and left cystic ovary. There was vaginal bleeding 13 days after operation for 9 days. Past history was irrelevant, menstrual cycle 12×27-30×4; one full term pregnancy. Six weeks following operation menopausal symptoms developed. See figure 1.

Experimental procedures were started 11-13-33. In all cases the days, rather than the dates are used in the figures, e.g., November 13, 16, 20, 23, 27, correspond to 1, 4, 8, 11 and 15 of the first period, figure 1. On the 1st, 4th, 8th, 11th and 15th days the patient was given 10,000 R. U. of estrin. Bleeding started on the 23rd day, 8 days after the last injection, and lasted 3 days, requiring 12 napkins. The hot flashes ceased 3 days after the second injection and returned 1 day before the

onset of bleeding. They increased in number and severity until stopped by the estrin used in the next period.

The second period was started 12-30-33. A biopsy (B 1) was taken before the first injection and was typically atrophic. On the 1st, 2nd, 3rd, 10th and 14th

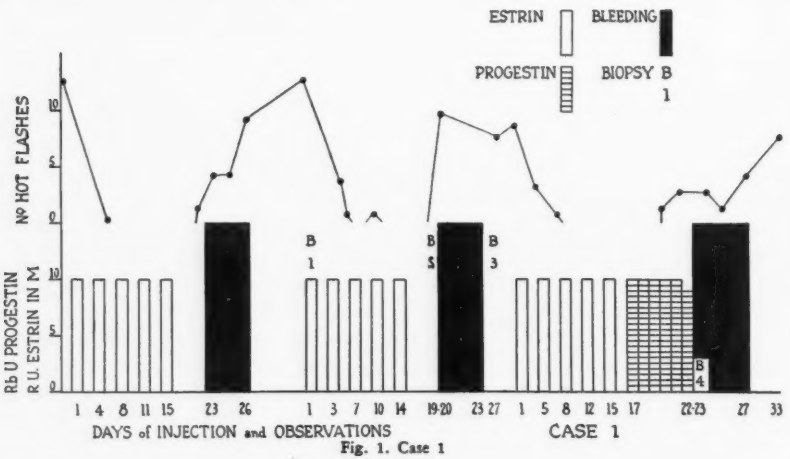


Fig. 1. Case 1

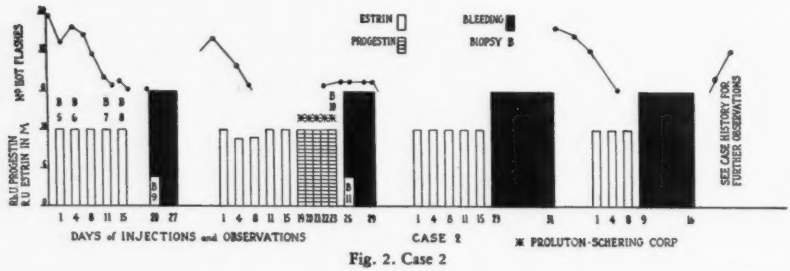


Fig. 2. Case 2

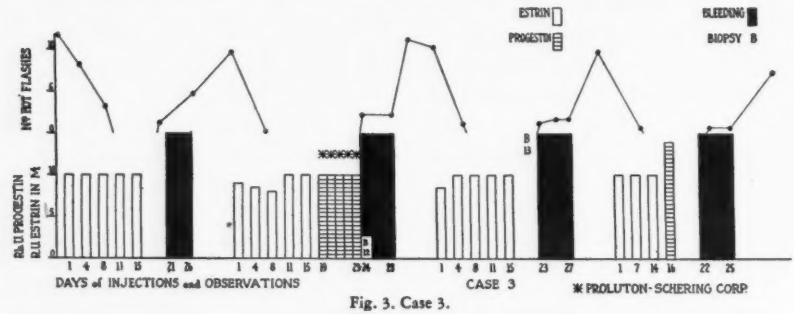


Fig. 3. Case 3.

days the patient was given 10,000 R. U. of estrin. A biopsy (B 2 and fig. 4) taken on the 19th day was mid-interval in type and the stroma was quite hemorrhagic. Bleeding started on the twentieth day, 6 days after the last injection, and lasted 4 days. The hot flashes were relieved after the second injection, and returned one day

before the onset of bleeding. A biopsy (B 3) taken 4 days after bleeding ceased, was characteristically resting. Only a little tissue could be obtained.

The third period was started 1-25-34. On the 1st, 5th, 8th, 12th and 15th days 10,000 R. U. of estrin was given. On the 17th, 18th, 19th, 20th, 21st and 22nd days 10 Rb. U. of progestin was given. A biopsy (B 4 and fig. 4) taken on the 23rd

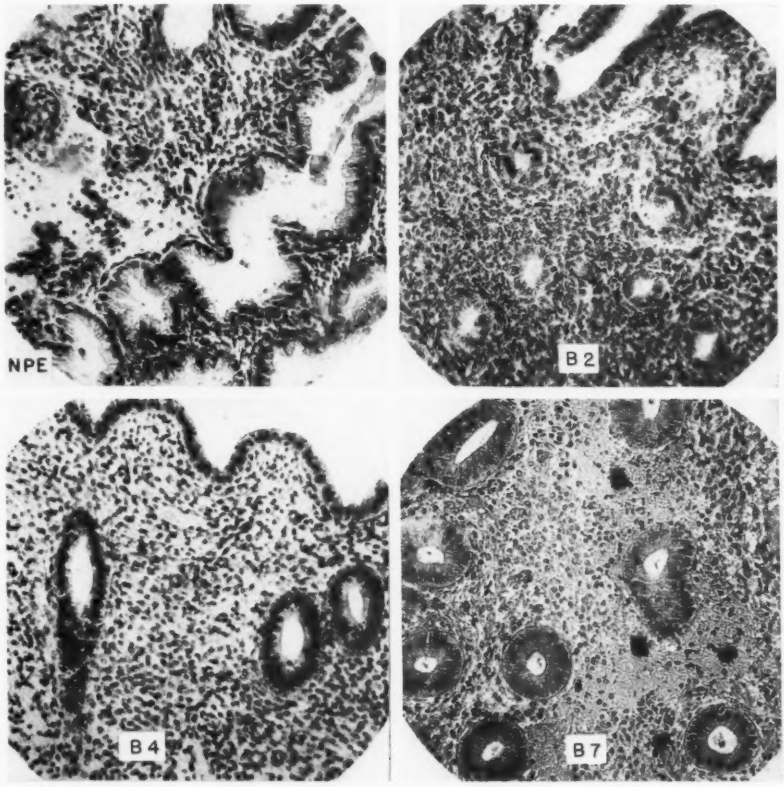


Fig. 4. Photomicrographs of endometrium. ×126.

NPE, normal human premenstrual endometrium, obtained about 12 hours before the period. The glands are much more dilated and tortuous than those shown by the other biopsies. There is much less hemorrhage than in the endometrium prepared by estrin alone. Shown for comparison with biopsies.

B 2. The endometrium is about the thickness of mid-interval type. The glands are about normal size; the nuclei of the gland cells are basilar. The stroma is hemorrhagic and shows some signs of disintegration. The surface epithelium is mid-interval in type.

B 4. This sample of endometrium was taken after estrin and progestin therapy and just before the onset of bleeding. The histological picture is essentially the same as that in biopsy 2 except that the stroma is a little more hemorrhagic and the glands a little more dilated.

B 7. This sample was taken about 3 days after the third injection of estrin. The picture is not unlike that of normal mid-interval endometrium.

day was fixed in Zenker's solution for the usual hematoxylin-eosin stain, and in absolute alcohol for glycogen staining. The endometrium was mid-interval in type and the stroma very hemorrhagic. The sample fixed in alcohol and stained with Best's carmine showed small glycogen granules diffusely scattered through the cells. The amount of glycogen was not more than normally present at this stage of development (6).

Bleeding started on the 23rd day, about 12 hours after the biopsy, and 8 days after the last injection of estrin. It lasted 4 days and was more profuse than with estrin alone. The hot flashes returned 2 days before bleeding started and gradually increased in number and severity until they were as frequent and severe as originally.

Case 2. A 19 year old, single female had a bilateral salpingo-oophorectomy, 11-2-33, for bilateral ovarian cystomas, with probable pyosalpinx. Many adhesions were encountered, past history irrelevant, menstrual cycle $12\times28\times3$. Menopausal symptoms developed 6 weeks following operation. See figure 2.

Experimental procedures were started 2-5-34. On the 1st, 4th, 8th, 11th and 15th days the patient was given 10,000 R. U. of estrin. Bleeding started the twentieth day, 5 days after the last injection, and lasted 7 days. Biopsies were taken on the 1st, 4th, 11th and 15th days. The biopsy (B 7 and fig. 4) was mid-interval in

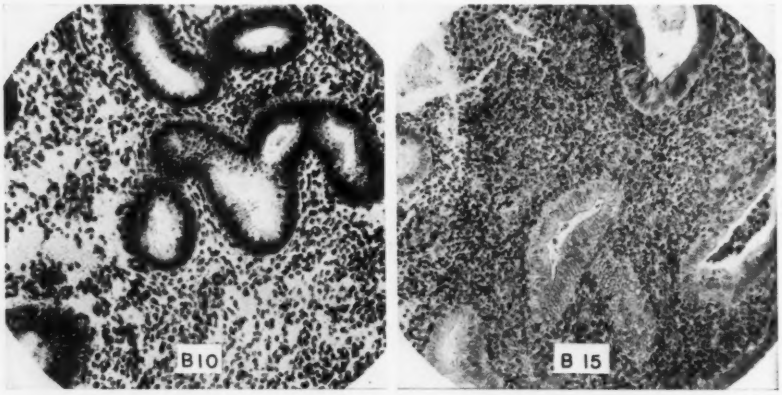


Fig. 5.
B. 10. This sample of endometrium taken after 50,000 R. U. of estrin and 50 Rb. U. of Proluton (progestin) and 2 days before the onset of bleeding shows no premenstrual change. The histological picture is similar to biopsy 4 except that there is less glycogen present.
B 15. The histological picture is that of mid-interval endometrium. The sample was taken 1 day before bleeding and shows no evidence of premenstrual change.

type with a small amount of hemorrhage into the stroma. The hot flashes were not entirely relieved until the last injection, and returned 1 day before the onset of bleeding.

The second period was started 3-6-34. On the 1st, 4th, 8th, 11th and 15th days 10,000 R. U. of estrin was given, and on the 19th, 20th, 21st, 22nd and 23rd days 10 Rb. U. of Proluton (progestin) was given. Bleeding started on the 25th day, 10 days after the last injection of estrin, lasted 4 days and was normal in amount. A biopsy (B 10 and fig. 5) taken on the 23rd day, part fixed in Zenker's solution and part in absolute alcohol, showed no proliferative changes or increase in glycogen. The hot flashes ceased 3 days after the second injection of estrin and returned 2 days before the onset of bleeding.

The third period was started 4-10-34. On the 1st, 4th, 8th, 11th and 15th days 10,000 R. U. of estrin was given. Bleeding started the 23rd day, 8 days after the last injection, and lasted 9 days.

The observations on this patient for the next year are perplexing. The fourth period was started 5-18-34. On the 1st, 4th and 8th days 10,000 R. U. of estrin was given. Bleeding started on the ninth day, lasted 6 days, and was very profuse,

requiring 3 dozen napkins. Hot flashes returned with the onset of bleeding and continued for some time afterward. Spontaneous bleeding occurred on the following dates: June 20-26, July 24-29, October 2-8, October 14, November 5-14, December 8-14, December 23-31, January 22-February 2. There was no bleeding in August or September, and there has been none since January 1935. Hot flashes during the last year have been frequent and severe in spite of the spontaneous bleeding. Many biopsies taken during this time showed only a resting endometrium. The patient has gained 50 pounds in weight. The spontaneous bleeding, gain in weight and severe hot flashes, all without medication, are paradoxical.

Case 3. A 33 year old widow had a right salpingo-oophorectomy in 1923 for an ectopic pregnancy. On 12-11-33 a left salpingo-oophorectomy for an ovarian cyst was done, and no adhesions were encountered. The past history was irrelevant, menstrual cycle 12 × 28 × 6, with severe dysmenorrhea since the onset of menstruation. The pain always started a few days before catamenia, continued until the menstrual flow was well established, and often required sedation. Hot flashes developed 6 weeks after operation.

Experimental procedures were started 1-26-34. See figure 3. On the 1st, 4th, 8th, 11th and 15th days 10,000 R. U. of estrin was given. Bleeding started on the 21st day, 6 days after the last injection, and lasted 6 days. The hot flashes abated after the third injection and returned 1 day before the onset of bleeding. Dysmenorrhea started on the 14th day, the pains increased in severity aand subsided after bleeding started. They were described by the patient as, "just like they used to be all my life."

The second period was started 3-6-34. On the 1st, 4th, 8th, 11th and 15th days 10,000 R. U. of estrin was given. On the 19th, 20th, 21st, 22nd and 23rd days 10 Rb. U. of Proluton (progestin) was given. A biopsy (B 12) taken on the 24th day was mid-interval in type and showed no progestin effect. Bleeding started on the 24th day, 9 days after the last injection of estrin, and lasted 4 days. The hot flashes stopped after the third injection of estrin and returned the day of bleeding. There was no dysmenorrhea this period.

The third and fourth periods, not shown in figure 3, were started 4-10-34 and 5-17-34 respectively. In each case on the 1st, 4th, 8th, 11th and 15th days 10,000 R. U. of estrin was given. Bleeding occurred 8 days after the last injection. Dysmenorrhea was severe in both cases.

The fifth period was started 6-28-34. On the 1st, 4th, 8th, 11th and 15th days 10,000 R. U. of estrin was given. A biopsy (B 13) taken on the twentieth day was mid-interval in type with considerable hemorrhage in the stroma. Bleeding occurred on the 23rd day, 8 days after the last injection, and lasted 5 days. The hot flashes returned the day of bleeding. Dysmenorrhea was marked.

The sixth period was started 10-12-34. Because of the previous observation (case 2), that the maximum growth of endometrium without excess hemorrhage, was produced with 30,000 R. U. of estrin, this patient was given 10,000 R. U. on the 1st, 7th and 14th days. On the sixteenth day 14 Rb. U. of progestin was given, and a local reaction developed almost immediately so that it was deemed unwise to continue progestin. Unexpected bleeding began on the 22nd day, 8 days after the last injection of estrin, and continued 3 days.

Case 4. A 49 year old, single nullipara, had a bilateral salpingo-oophorectomy 9-16-34, for a large twisted ovarian cyst on the right, and a small cystic ovary on the left. The past history was irrelevant, menstrual cycle 14 × 28 ×5.

Based on the observations in case 3, period 6, this patient was given a total of 30,000 R. U. of estrin followed by progestin. See figure 6. Injections were started

1-25-35. On the 1st, 7th and 14th days 10,000 R. U. of estrin was given. On the fifteenth day 14 Rb. U. of progestin was given. In spite of a purer preparation, a marked local reaction developed. A biopsy (B 14) taken on the seventeenth day showed no premenstrual changes. Bleeding started on the 19th day, 5 days after the last injection of estrin and lasted for 6 days. The hot flashes which abated after the second injection of estrin, returned after the bleeding was over.

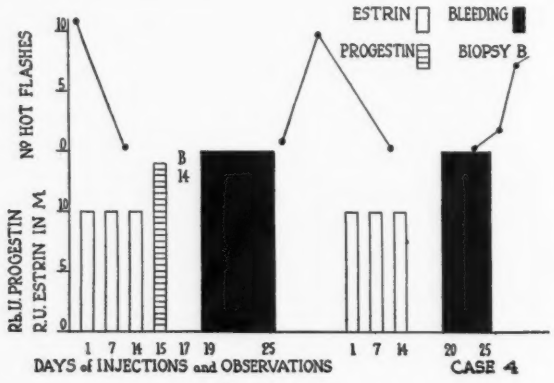


Fig. 6. Case 4

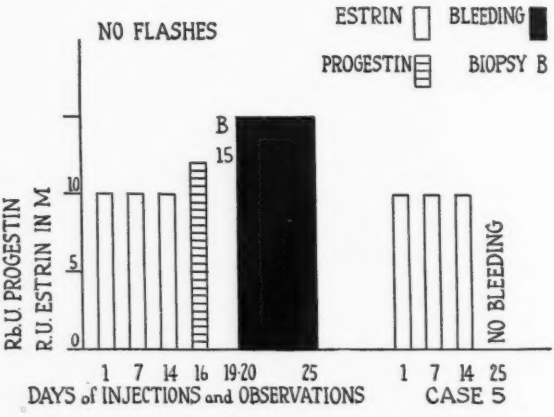


Fig. 7. Case 5.

A second period was started 3-25-35. On the 1st, 7th and 14th days 10,000 R. U. of estrin was given. Bleeding started on the twentieth day, 6 days after the last injection, and continued for 5 days. The hot flashes ceased after the second injection and returned after 3 days bleeding.

Case 5. A 31 year old, divorced female, had a left salpingo-oophorectomy at another hospital 6-22-33. On 3-5-34 a right oophorectomy was done for a cystic ovary. The past history was irrelevant, menstrual cycle 12×28×5-6. Since her last operation there have been only a few mild hot flashes which were entirely relieved by therapy or time.

Observations were started 2-27-35. See figure 7. On the 1st, 7th and 14th days 10,000 R. U. of estrin was given. On the sixteenth day 12 Rb. U. of progestin was given. A biopsy (B 15 and fig. 5) was taken on the nineteenth day and showed a mid-interval endometrium with no premenstrual or progestin changes. Bleeding started on the twentieth day, 6 days after the last injection of estrin, and continued for 6 days.

A second period was started 3-25-35. On the 1st, 7th and 14th days 10,000 R. U. of estrin was given. No bleeding followed.

DISCUSSION

From the foregoing observations it has been shown, first: that premenstrual endometrium cannot be prepared in the castrated human female with the amounts of the two hormones used. The samples of endometrium (B 4, 10, 12) taken during these observations after progestin have shown no typical premenstrual changes, according to the standards of Bartlemez and Bensley (6). Neustaedter (7) was unable to produce a premenstrual endometrium in a human castrate by giving 86,400 R. U. of estrin by mouth over a period of 24 days followed by 10 Rb. U. of luteo-hormone for 6 days. This may be due to several factors, such as excess estrin, deficient progestin or lack of balance between these two hormones. Hisaw (8) has recently produced proliferative changes in the mucosa of two castrated juvenile monkeys with corporin (progestin) alone. The effects, he states, are limited mostly to the structures existing before treatment and differ from that produced by estrin with corporin (progestin), in that estrin furnishes the structures on which progestin can act.

In the present work 30,000 R. U. of estrin produced a typical mid-interval type of endometrium (B 7, fig. 4). Larger doses produced considerable hemorrhage (B 8, 9) in the stroma and early degenerative changes. Bleeding was produced by 30,000 R. U. of estrin if followed by 12-14 Rb. U. of progestin, but no premenstrual changes were noted (B 14, 15) in the biopsies of endometrium. Before further observations can be made in the human it will be necessary to crystallize progestin, since the crude preparations cause marked local reactions.

Second: Bleeding can be produced by 50,000 R. U. of estrin alone, less being ineffective, (9). Bleeding thus produced starts, on the average, 7 days after the last injection. When estrin is followed by progestin the onset of bleeding is delayed on the average, only 2 days. Contrast this with the marked 28 day delay produced in the castrated female monkey (10), by giving the equivalent of 70 grams of fresh corpus luteum daily for 28 days.

Third: Menopausal symptoms, particularly hot flashes, are only temporarily relieved by estrin when given in sufficient amounts. The symptoms usually return one day before the onset of bleeding and gradually increase in frequency and severity. In each case after experimentation was stopped the hot flashes were as frequent and severe as originally. Apparently estrin brings about a delay of the normal adjustment of menopause by its inhibitory action on the pituitary gland. If this is the case one must look elsewhere for the etiology of the symptom complex. It is highly probable that the pituitary gland is involved.

SUMMARY

A premenstrual endometrium could not be prepared in five castrated human females using 50,000 R. U. of estrin and 12-60 Rb. U. of progestin. No premenstrual changes were noted when 30,000 R.U. of estrin and 12-14 Rb. U. of progestin were given. Bleeding can be induced in the castrated human female with 50,000 R. U. of estrin alone. It is not markedly delayed if estrin plus progestin is given. Bleeding can be produced with only 30,000 R. U. of estrin if followed by 12-14 Rb. U. of progestin. Hot flashes are only temporarily relieved with estrin. When therapy is stopped the symptoms return in their original frequency and severity.

REFERENCES

1. Allen, W. M.: J. Biol. Chem. **98:** 591. 1932.
2. Allen, W. M., and R. K. Meyer: Am. J. Physiol. **106:** 55. 1932.
3. Allen, W. M., and G. W. Corner: Am. J. Physiol. **88:** 340. 1929.
4. Kaufmann, C.: Zentralb. f. Gynäk. **56:** 2058. 1932.
5. Kaufmann, C.: Klin. Wchnschrft. **40:** 1. 1933.
6. Bartlemez, G. W., and C. M. Bensley: Special Cytology. Cowdry, **3:** 1932.
7. Neustaedter, T.: Am. J. Obst. & Gynec. **29:** 680. 1935.
8. Hisaw, F. L.: Am. J. Obst. & Gynec. **29:** 638. 1935.
9. Elden, C. A.: Unpublished data.
10. Smith, P. E., and E. T. Engle: Proc. Soc. Exper. Biol. & Med. **29:** 1225. 1932.

THE CALORIGENIC ACTION OF EXTRACTS OF THE ANTERIOR LOBE OF THE PITUITARY IN MAN[1]

W. O. THOMPSON, S. G. TAYLOR, III,[2] P. K. THOMPSON, S. B. NADLER, AND L. F. N. DICKIE

From the Department of Medicine, Rush Medical College of the University of Chicago, the Presbyterian and Cook County Hospitals

CHICAGO, ILLINOIS

and

The Department of Biochemistry, Tulane University

NEW ORLEANS, LOUISIANA

The present series of observations represents an attempt to reproduce in man results which have been observed in animals during the administration of extracts of the anterior lobe of the pituitary containing the so-called thyrotropic principle.[3] Abundant evidence of the relationship between the pituitary and the thyroid has been obtained in animals and in man and has been summarized in recent articles by Evans (3), (4), Smith (5) and Collip (6), (7). The following facts have been demonstrated.

1. Enlargement of the pituitary has been noted in thyroidectomized animals and in patients with cretinism and myxedema.

2. The thyroid becomes atrophic and the basal metabolism drops following removal of the pituitary.

3. The administration of active pituitary extracts to animals causes, temporarily: (a) hyperplasia of the thyroid, (b) reduction in the iodine content of the gland, (c) increase in the organic iodine of the blood, (d) tachycardia, (e) exophthalmos, (f) increase in basal metabolic rate. These changes do not occur in thyroidectomized animals.

4. In cases of hypopituitarism in man, the basal metabolism is low and the thyroid atrophic. Conversely, in the early stages of acromegaly a goiter is commonly found associated with a high basal metabolism.

5. Iodine is effective in reducing the high basal metabolism of patients with acromegaly and of animals which are receiving pituitary extracts.

6. The metabolic and thyroid changes produced by pituitary extracts are always temporary and prolonged administration in animals may cause the metabolism to drop to as low a level as that seen after hypophysectomy. The serum of such animals contains some factor which inhibits the action of the thyrotropic principle. This observation has given rise to the anti-hormone theory of Collip (8).

While the calorigenic action of pituitary extracts has been conclusively demonstrated in animals, it has been observed in only isolated instances in man and it seemed desirable to observe the effect of active extracts in

[1] Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 10, 1935.

[2] Squibb Research Fellow.

[3] Brief summaries of part of these data have been reported by Thompson, Thompson, Taylor, Nadler and Dickie (1), (2).

TABLE 1.

Date treatment started	Patient	Sex and age	Level of B.M.R. before treatment	Level to which B.M.R. rose during treatment	No. of points rise (if 10 or more)	Day rise began	Days required to reach maximum	Day B.M.R. began to drop	Level of B.M.R. at end of treatment	Medication, to time of last B.M.R.			
										Usual daily dose cc.	Total cc. administered	No. days administered	
MARKED HYPOTHYROIDISM													
9/27/33	G.H.	M 53	−46	−46					−46	4	84	20	Squibb's Ant. Pit. Ext. S.C.
6/14/34	B.L.	F 32	−38	−35					−35	8	126	19	" " " " "
7/ 6/34			−35	−31					−36	6	176	25	Phyone S.C.
3/29/34	R.R.	F 34	−27	−25					−26	1.5	14	18	" "
6/ 5/34	G.W.	F 41	−38	−32					−38	4–6	120	22	Squibb's Ant. Pit. Ext. S.C.
MILD OR MODERATE HYPOTHYROIDISM (A) WITHOUT GOITER													
3/27/34	O.B.	F 52	−15	−13					−15	4–8	118	21	" " " " "
4/17/34			−15	+ 2	17	4– 6	13–15	18–20	−15	8	137	34	Phyone S.C.
5/24/34			−15	−16					−17	8	40	14	Squibb's Ant. Pit. Ext. S.C.
2/ 2/35	A.B.	M 45	−20	−13					−26	2–6	225	42	" " " " "
4/ 5/34	L.H.	F 46	−13	+25	38	8–13	15–20	26–34	−16	4	250	84	Phyone S.C.
3/13/34	F.M.	F 29	−18	+24	42	1– 6	15	21–22	+18	4	92	24	" "
10/26/34	E.M.	F 32	−15	− 9					−17	2–4	48	14	Phyone Heated S.C.
11/11/34			−17	− 8					−17	4	30	8	Phyone S.C.
7/ 6/34	R.P.	M 25	−13	+15	28	1– 3	11–12	16–19	−19 then −5	3	183	71	" "
6/12/34	G.W.	F 42	−24	−25					−25	8	165	28	Squibb's Ant. Pit. Ext. S.C.
9/ 4/34	A.C.	F 24	−22	− 5	17	1– 3	6– 8	9–11	−20	3–4	55	20	Phyone S.C.
3/14/35			−20	−18					−18	4–5	79	27	Protein-free Phyone S.C.
5/ 4/35	C.B.	M 41	−19	−19					−19	6	132	30	" " " "

(B) WITH GOITER													
12/23/33	G.D.	F 24	−26	+12	38	1– 2	12–13	14–15	+ 3	8	87	16	Phyone S.C.
2/ 2/34			−26	−22					−28	4–8	74	14	" "
5/11/34	M.J.	F 42	−29	+17	46	1– 2	9–10	11	+ 5	8	122	18	Squibb's Ant. Pit. Ext. S.C.
7/11/34			−21	−25					−25	8	36	5	" " " " "
6/ 6/34	A.C.	F 34	−23	−22					−24	2–8	34	16	" " " " "
7/ 6/34			−24	−21					−21	4–6	50	14	Phyone S.C.
5/25/34	E.W.	F 50	−24	− 5	19				− 5	4	16	6	Squibb's Ant. Pit. Ext. S.C.
HYPOPITUITARISM													
12/13/33	J.R.	M 17	−14	− 4	10	8	8		− 4	4– 8	106	25	Squibb's Ant. Pit. Ext. S.C.
12/20/33	T.H.	M 14	−24	−10	14	4	5– 6	19	−19	4–16	168	25	" " " " "
1/15/34			−19	−22					−24	4	41	11	Phyone S.C.
3/23/35	D.C.	M 14	−18	− 8	10	3– 4	3– 4	18–19	−22	4– 6	260	60	Protein-free Phyone S.C.
8/23/33	F.C.	F 17	−26	−23					−26	2	24	14	Squibb's Ant. Pit. Ext. S.C.
2/ 4/34	A.M.	M 50	−30	+ 6	36	4	22	42–43	−27	4	419	106	Phyone S.C.
12/ 1/34			−20	−11					−21	3	95	56	" "
5/19/34	B.M.	F 41	−28	−12	16	6– 7	8– 9		−12	4	51	10	Squibb's Ant. Pit. Ext. S.C.
6/18/34			−29	−24					−24	8	358	48	" "
6/ 1/34	M.S.	F 45	−12	−23					−23	20	373	27	Phyone by mouth.
1/18/35			− 7	− 5					−15	6	225	90	Phyone S.C.
3/14/35	J.H.	M 15	−15	+13	28	1– 9	10–16		+13	3	38	16	Protein-free Phyone S.C.
3/15/35	F.K.	M 17	−18	−15					−15	4– 6	129	96	" " " "
4/10/35	W.R.	F 35	−19	− 4	15	9–10	9–10	23	−22	6	130	55	" " " "
UNCLASSIFIED: LOW BASAL METABOLISM													
6/19/34	P.A.	M 15	−25	−11	14	29	34	37–39	−19	8	316	44	Squibb's Ant. Pit. Ext. S.C.
6/ 5/34	M.L.	F 31	−18	−17					−17	20	286	21	Phyone by mouth.
3/30/34	L.S.	F 52	−19	− 4	15	1– 7	11–14	15–21	−17	4	107	35	Phyone S.C.
4/28/34	R.F.	F 16	−18	+ 8	26	1– 7	—	—	−18	4	110	49	" "
3/ 7/35			−15	−17					−17	4–6	145	48	Protein-free Phyone S.C.

TABLE 1 (*Continued*)

Date treatment started	Patient	Sex and Age	Level of B.M.R. before treatment	Level to which B.M.R. rose during treatment	No. of points rise (if 10 or more)	Day rise began	Days required to reach maximum	Day B.M.R. began to drop	Level of B.M.R. at end of treatment	Medication, to time of last B.M.R.			
										Usual daily dose cc.	Total cc. administered	No. days administered	
WITH OBESITY													
1/28/35	M.D.	F 15	−13	−15					−15	2	16	9	Acetone-precipitate from Protein-free Phyone S.C.
2/ 6/35			−15	−18					−18	1–5	167	70	Phyone S.C.
2/ 4/34	A.N.	F 40	−12	+15	27	1– 5	14–15	28–29	−23 then −14	4–8	376	108	" "
5/25/34			−14	−10					−10	8	40	12	Squibb's Ant. Pit. Ext. S.C.
WITH GOITER													
12/12/33	S.S.	F 36	−12	−15					−15	3–4	65	27	" " " " "
WITH DIABETES MELLITUS													
12/ 1/34	C.B.	M 17	−22	−21					−21	2	18	9	Phyone S.C.
2/23/34	S.M.	M 16	−19	−21					−20	4	320	90	Squibb's Ant. Pit. Ext. S.C.
NORMAL BASAL METABOLISM WITH OBESITY													
3/13/34	V.A.	F 24	+2	+13	11	1–15	—	16–23	−6	4	95	50	Phyone S.C.
5/18/34	A.B.	F 37	−1	+13	14	5	5	14	+1	8	172	34	Squibb's Ant. Pit. Ext. S.C.
10/25/34	M.K.	F 37	−8	+ 1					±0	6	304	53	Phyone S.C.
3/28/35	L.H.	F 34	+2	± 0					±0	2–3	46	24	Protein-free Phyone S.C.

With Goiter													
12/12/33	C.D.	F 18	+2	+21	19	5–10	5–10	11–14	+ 1	4–8	123	43	Squibb's Ant. Pit. Ext. S.C.
3/19/34			±0	± 0					± 0	4	134	43	Phyone S.C.
5/19/34	V.M.	F 35	+7	+17	10	7– 9	7– 9	14	+16	4–8	68	12	Squibb's Ant. Pit. Ext. S.C.
3/30/34	N.C.	M 46	−3	+28	31	1– 3	7	31–34	− 9	4	262	65	Phyone S.C.
5/18/34	C.F.	F 41	−5	+ 1					+ 1	8	136	34	Squibb's Ant. Pit. Ext. S.C.
3/23/34	L.M.	F 29	+7	+13					− 5	4	142	64	Phyone S.C.
12/13/33	A.T.	F 27	−3	− 5					− 5	4	22	10	Squibb's Ant. Pit. Ext. S.C.
Miscellaneous													
5/16/34	M.T.	M 44	+ 9	+11					+12	8	340	62	Squibb's Ant. Pit. Ext. S.C.
1/ 2/34	J.P.	F 18	+17	+17					+17	4	44	17	" " " " "
Thyrotoxicosis													
7/19/34	G.H.	M 40	+58	+68	10	1– 3	1– 3	—	+68	3	64	30	Phyone S.C.
8/13/34	B.J.	M 31	+45	+56	11	26	26	41–42	+37	5–8	437	85	Squibb's Ant. Pit. Ext. S.C.
2/ 7/35	R.J.	M 35	+46	+53					+53	1.5	47	55	Phyone S.C.
7/12/34	J.K.	F 43	+21	+37	16	1– 5	5– 7	10–11	+15	8	504	88	Squibb's Ant. Pit. Ext. S.C.
11/21/34			+19	+31	12	12	26	32–37	+19	4	252	91	" " " " "
1/18/35	S.L.	F 20	+28	+12					+12	5	224	138	Phyone S.C.
4/18/34	J.M.	M 44	+32	+50	18	31	33	—	+50	4	154	40	Squibb's Ant. Pit. Ext. S.C.
6/22/34			+23	+41	18	1	1	9–10	+15	8	620	137	" " " " "
5/15/35			+ 1	+ 9					+ 9	4	76	22	" " " " "
6/ 4/34	E.R.	F 37	+16	+16					+16	4–6	93	20	" " " " "
4/15/35	E.V.	F 43	+49	+62	13	1–8	1– 8	16–33	+31	5	50	33	Phyone S.C.
1/25/35	J.S.	M 43	+45	+42					+42	3	28	12	Acetone-precipitate from Protein-free Phyone S.C.
2/ 6/35			+43	+51					+42	4	154	72	Phyone S.C.
2/28/35	J.A.	M 27	+46	+52					+52	4	55	15	Protein-free Phyone S.C.
2/28/35	N.R.	F 35	+55	+68	13	1– 4	1–4	—	+68	3	20	15	" " " "

patients with various degrees of thyroid function and in particular to observe the effect of prolonged administration.

METHOD

We have used four types of extract. 1) The anterior pituitary extract of Squibb's,[4] prepared by a modification of the method of Van Dyke and Wallen-Lawrence. This is an alkaline aqueous extract containing the growth, thyrotropic and sex complementary factors of the anterior pituitary. 2) Phyone of the Wilson Laboratories,[5] an alkaline extract of the anterior lobe, prepared according to the method of Van Dyke and Wallen-Lawrence (9) and assayed according to their procedure. In addition to the growth hormone, it contains some gonadotropic hormone, some thyrotropic factor and some prolactin. 3) A protein-free phyone, obtained by precipitating the protein from phyone with trichloracetic acid. In 2 instances we used the material precipitated from the protein-free phyone by treatment with 10 volumes of acetone. 4) Heated phyone was used in one instance. All material was administered subcutaneously with the exception of two observations with phyone administered by the oral route.

DATA

A total of 79 observations have been made on 59 patients (tables 1 and 2). These may be grouped as follows.

Marked hypothyroidism: 5 observations on 4 patients.

Mild or moderate hypothyroidism:

a) Without goiter, 13 observations on 9 patients.

b) With goiter, 7 observations on 4 patients.

Hypopituitarism: 14 observations on 10 patients.

Unclassified low basal metabolism: 12 observations on 9 patients;
2 had diabetes, 2 were obese and 1 had goiter.

Normal basal metabolism: 11 observations on 10 patients;
4 were obese and 6 had goiter.

Miscellaneous: 2 observations on 2 patients.

Thyrotoxicosis: 15 observations on 11 patients.

It may be seen that phyone and Squibb's extract each produced an increase in metabolism in about 50 per cent of the patients when given subcutaneously, and protein-free phyone in about one-third of the patients. In the two patients in whom the acetone-precipitated material was used, no response was obtained. Phyone was ineffective in the two instances in which it was given by mouth. Heated phyone was without effect in one patient by the subcutaneous route.

The increase in metabolism varied from 10 points to 46 points (an increase in metabolism of less than 10 points was not considered a rise), the average increase being 17 points. The rise usually began about the fifth day, although sometimes much later, after treatment was started and the maximum duration of the increase was about two months.

[4] This material was donated by E. R. Squibb and Sons through the kindness of Dr. John F. Anderson and Dr. J. A. Morrell.

[5] This material was donated by the Wilson Laboratories, Chicago, through the kindness of Dr. D. Klein.

In 18 patients two or more series of observations were made. In 11 of these the first course of treatment was associated with an increase in metabolism, but in only two instances was the second course accompanied by an increase. In another instance no increase occurred during the administration of the Squibb product but did occur shortly afterward during the administration of phyone. In one of these three instances the first series of injections had not been continued until the metabolism had returned to its original level or lower. In two cases, no rise occurred during the second course even when the first series of injections had been discontinued before the metabolism had returned to its preinjection level. In nine instances, during prolonged treatment, the metabolism finally fell to a lower level than it was initially.

TABLE 2.

Medication	Method of Administration	No. of Patients	No. of observations	On basis of number of observations	
				Increase in B.M.R. of 10 points or more	No increase in B.M.R.
Squibb's anterior pituitary extract	Subcutaneously	28	34	14	20
Whole phyone	Subcutaneously	28	30	14	16
Whole phyone	By mouth	2	2	0	2
Heated phyone	Subcutaneously	1	1	0	1
Protein-free phyone	Subcutaneously	12	12	4	8

In one of these patients this reduction was not preceded by a rise in metabolism. In one patient no effect was observed either from heated phyone or from whole phyone given immediately afterward.

Of thirteen patients whose sera were tested by Dr. Ludvig Hektoen, the development of precipitins was noted only in one.

A résumé of the data shows that the only group of patients in which no response was observed was that with marked myxedema. However, objections to the data on these patients are that the treatment was not continued as long as in many of the others and the number of cases (four) was small. If they reflect the true situation, it would appear that as long as there is any thyroid tissue capable of functioning it can be driven temporarily to increased activity by suitable pituitary extracts. Thus, in patients with moderate hypothyroidism and basal metabolic rates as low as minus 25 per cent, the metabolism rose in about as large a percentage as in patients with normal or high metabolic rates. The rise in patients with pituitary tumors perhaps suggests that the pituitary failure in these cases was not the result of the development of inhibiting substances, but rather the effect of pituitary failure per se. In the light of these observations the presence of a low basal metabolism in patients with goiters of varying size takes on added interest. The effect of the pituitary extracts would indicate that under a suitable stimulus these goiters are capable of supplying enough of the thyroid hormone to raise the rate to normal. Their failure to do so may perhaps be the result of inade-

quate stimulation of the thyroid. Of equal interest perhaps is the production of a temporary period of hyperthyroidism in patients with non-toxic goiters and normal levels of metabolism, and the production of transient periods of increased severity in patients with exophthalmic goiter. Fifteen observations were made on eleven patients with exophthalmic goiter. In eight instances the metabolism rose ten or more points and in seven no increase was noted. Of the seven instances in which there was no rise, one followed two previous courses and one, one previous course of treatment. Of the eight rises, two occurred during second courses of treatment and one of these two after the metabolism had returned to its preinjection level during the first course. Five patients failed to show a significant increase in metabolism. Of seven patients who received the extract for 35 days or longer, in three no significant change was noted; in three the metabolism after an initial increase, fell to a lower level than before treatment was started and in one case began to fall without an initial increase. In two patients the rate fell to within normal limits during the administration of or shortly after omission of the extract. It is obvious that more data are necessary to determine what rôle, if any, the pituitary may play in toxic goiter and whether or not protective substances can be developed which will control the disease.

The patients who showed no response are fully as interesting as those who responded. If Collip's conception of an antithyrotropic substance be correct, the question would naturally arise as to whether such a substance was present in these patients before the injections were started.

Observations in several patients have clearly shown that thyroxine and desiccated thyroid exert a well-marked calorigenic action after response to the pituitary extract has ceased, but whether or not it is altered in any way can not be said at present.

These observations serve to emphasize what has previously been suggested, namely, that in all thyroid disturbances the rôle of the pituitary must be considered. It is possible that disorders which we have come to regard as purely thyroid in origin may prove to be caused either from lack of adequate stimulation or overstimulation of the thyroid. However, conclusions must be drawn with caution and it will take several years to define more precisely the rôle of the pituitary in thyroid disorders.

SUMMARY AND CONCLUSIONS

In 32 of 79 observations on 59 patients of various types, an increase in basal metabolism was noted during the administration of pituitary extracts containing the thyrotropic principle. This increase occurred in every group of patients in whom there was any thyroid tissue capable of functioning. No rise was noted in 4 patients with marked myxedema, but in patients with mild or moderate hypothyroidism the rate could be raised to normal, patients with non-toxic goiters could be made toxic, and patients with exophthalmic goiter could be made worse.

The increase in metabolism was always temporary in spite of prolonged treatment, the metabolism eventually returning to its level before treatment or

even to a lower level. Second courses of treatment usually, but not invariably, failed to produce any increase in metabolism.

In 6 of 11 patients with exophthalmic goiter the severity of the disease was increased, while in 5 no definite change was noted. In 5 of the patients, the effect of prolonged treatment could not be observed because increase in the severity of the disease made it necessary to discontinue the extracts. In 3 the metabolism, following an initial rise, dropped to a lower level than it had been initially and in 1 patient whose metabolism had been stationary for a long time during the administration of iodine, no initial rise was noted, but the metabolism began to fall toward the normal level shortly after the administration was begun.

Thyroxine and desiccated thyroid produced well-marked calorigenic effects after patients had become refractory to the pituitary extracts.

These observations show that in disorders of thyroid function the possible rôle of the pituitary should be considered.

REFERENCES

1. Thompson, W. O., P. K. Thompson, S. G. Taylor, III, S. B. Nadler, and L. F. N. Dickie: Endocrinology **19:** 14. 1935.
2. Thompson, W. O., P. K. Thompson, S. G. Taylor, III, S. B. Nadler, and L. F. N. Dickie: J.A.M.A. **104:** 972. 1935.
3. Evans, H. M.: J.A.M.A. **104:** 464. 1935.
4. Evans, H. M.: J.A.M.A. **104:** 1232. 1935.
5. Smith, P. E.: J.A.M.A. **104:** 548. 1935.
6. Collip, J. B.: J.A.M.A. **104:** 916. 1935.
7. Collip, J. B., and E. M. Anderson: J.A.M.A. **104:** 865. 1935.
8. Collip, J. B., and E. M. Anderson: Lancet **1:** 76. 1934.
9. Van Dyke, H. B., and Z. Wallen-Lawrence: J. Pharmacol. & Exper. Therap. **40:** 413. 1930.

ENDOCRINE DWARFISM[1]

THIRD REPORT

ROBERT L. SCHAEFER
DETROIT, MICHIGAN

In two previous communications (1), (2) we have discussed the classification of endocrine dwarfism, its diagnosis, and the therapeutic results obtained in the first group of human beings to receive the pituitary growth hormone.[2] This publication is designed to show results obtained (a) in the original group of four persons who have been under treatment and observation for a period of about two years and nine months and (b) in a new group of seven persons who have been under treatment from six to nineteen months. Furthermore, metabolic observations before and during the administration of the growth hormone will be presented.

From table 1 it will be noted that treatment has been continuous in three of the four original patients. Patient 5 was withdrawn from treatment when roentgenographic studies for osseous development revealed epiphyseal closure and also because of the completion of sex maturity. For a time corresponding with his duration of treatment and age one could predict only 0.57 inches gain in height for the normally growing individual. The therapeutic result, as noted from a gain in statural height of 6.2 inches, is remarkable.

It is believed that the two clinical phenomena, epiphyseal closure and completion of sex maturity, are more than coincidental, and that epiphyseal closure is effected at the time of sex maturity through gonadal influence. Therefore, the administration of the pituitary growth hormone is clinically contra-indicated when epiphyseal closure occurs because of the probability of producing an abnormal type of growth such as is seen in adult acromegaly.

Chondro-epiphyseal changes, such as the moth-eaten, fuzzy, ragged and poorly outlined appearance of the osseous centers together with decreased bone density, at first noted in patients 3 and 4 have disappeared, and roentgenologically the structures are now normal. There has also been a definite advance in osseous development, which previously was retarded. Subjectively, the increase in appetite and physical activity has continued. Patient 7, during the course of treatment, developed a very definite neurohypophyseal obesity which was not manifest when treatment was begun. We do not believe that this is a result of medication but a condition occasionally observed in the pituitary dwarf.

The results in the other three patients who have continued treatment have been equally as good as in patient 5. According to Chart I, the average actual growth increment for the duration of treatment, 2.3 years, exceeds the average normal increment for respective ages and period of time by 3.1 inches. The results are even more striking if the three lines of growth are

[1] Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 10, 1935.
This work was instituted with Dr. Wm. Engelbach previous to his death.

[2] Antuitrin-G is a product not yet commercially available and was supplied to us by the Research Laboratory of Parke, Davis and Company for a preliminary research test. The letter "G" is used to differentiate the growth from the sex factor.

TABLE 1. ORIGINAL GROUP

Case	Before Treatment								After Treatment				
	Age		Actual Height	Normal Variation		Control Period	Gain in Height in Control Period	Osseous Development Delay	Duration of Treatment	Height	Normal Increment for Time Treated	Gain in Height	Remarks
	Yr.	Mo.	Inches	Min. In.	Max. In.	Months		Years	Months	Inches	Inches	Inches	
3 P.G. M −TP	11		47.5	53.3	57.1	3	None	4	33	56.5	5.0	9.0	Dosage of 5 cc. twice weekly for last eight months. Now approximates fairly normal boy of 14 yr. Advance in osseous development. Complete return to normal from chondro-epiphyseal changes first noted.
4 C.M. F −TP	9	5	42.7	50.1	53.7	2	None	3	33	49.2	6.1	6.5	Dosage of 5 cc. twice weekly for last nine months. Advance in osseous development. Return to normal from chondro-epiphyseal changes first noted in osseous studies. Improvement physically and mentally.
5 D.T. M −PT	18	6	52.5	64.9	69.3	5	0.5 inch	3	21	59.2	.57	6.2	Dosage of 2 cc. three times weekly throughout. Withdrawn from treatment because of epiphyseal closure and sex maturity.
7 R.S. F −P	7	4	36.7	45.2	48.4	6	None	1	27½	43.5	4.6	6.8	Dosage of 5 cc. twice weekly for last 3½ months. Developed neuro-hypophyseal obesity. Advance in osseous development.

−P denotes hypopituitarism. −TP denotes hypothyropituitarism. −PT denotes hypopituitarothyroidism. M, male; F, female.

TABLE 2. SECOND GROUP

Case	Before Treatment						After Treatment				
	Age		Actual Height	Normal Variation		Osseous Development Delay	Duration of Treatment	Height	Normal Increment for time treated	Gain in Height	Remarks
	Yr.	Mo.	Inches	Min. In.	Max. In.	Years	Months	Inches	Inches	Inches	
J.C. M —P	4	9	36.5	41.2	44.2	2–3	19	41.2	3.6	4.7	Dosage of 5 cc. twice weekly last 9 months. Not so timid nor nervous. Appetite increased.
J.D. M —PT	15	4	53.2	61.1	65.3	1–2	16	55.3	1.9	2.1	Dosage of 5 cc. twice weekly last 9 months.
L.W. M —PT	6		39.5	43.5	46.5	1–2	8	40.8	1.4	1.3	Dosage of 5 cc. twice weekly. General improvement.
D.B. M —P	3	3	29.7	37.4	40.2	1	11	30.9	2.5	1.2	Dosage of 5 cc. twice weekly last 2 months. General improvement.
J.B. M —P	9	3	37.8	50.4	54.0	2	11	39.7	1.8	1.9	Dosage of 5 cc. twice weekly last 9 months. General improvement.
B.B. M —P	12	6	39.8	56.0	60.0	1	11	41.7	1.6	1.9	Dosage of 5 cc. twice weekly last 9 months. General improvement.
M.P. F —P	13	10	52.4	59.3	63.5	Normal	6	54.6	.6	2.2	Dosage of 5 cc. twice weekly last 3 months.

—P denotes hypopituitarism. —PT denotes hypopituitarothyroidism.

compared. The actual growth obtained through treatment was 7.1 inches. The normal growth increment was 4 inches. The growth increase for these four individuals for a corresponding time previous to treatment, which is an assumption determined from their histories, former measurements, and actual heights at the time of initiation of treatment, was 1.9 inches.

The second group of patients (see table 2) were under observation and treatment from six months to one year and seven months. The patients were subjected to the same diagnostic routine undergone by the original four cases,

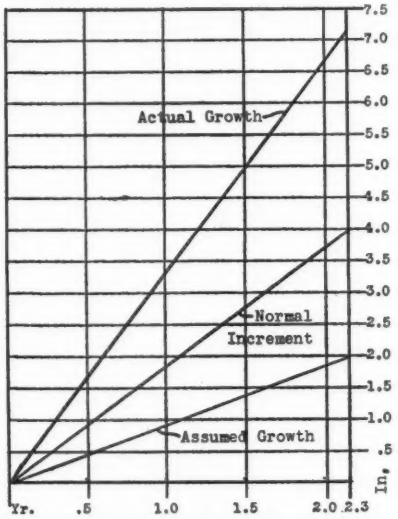


Chart I. Original group. Average duration of treatment, 2.3 years; average gain in height on treatment, 7.1 in.; average normal increment for age and time, 4.0 in.; average assumed growth for previous 2.3 years prior to treatment, 2.0 in.

consisting of a comprehensive history, physical examination, blood chemistry studies, basal metabolism, specific dynamic action, and roentgenographic studies for osseous development. Previous histories and former observations of mensuration, as far as could be ascertained, indicated the same inhibition of growth, and their actual measurements at the time of examination were far below the minimal normal. A control period was not deemed necessary after our experience with the original group. Only one of the original seven patients increased in height, and this was relatively negligible, being one-half inch in five months.

Diagnostically the second group displayed a definite delay in osseous development which we believe is a good prognostic omen. None of this series displayed the chondro-epiphyseal changes previously described. In addition to the treatment of the indicated dosage of the growth hormone a tolerant dose of thyroid was given, which we still believe is a valuable adjunct in treating pituitary dwarfism whether or not hypothyroid findings are demonstrated (3).

The second group of seven patients (Chart II) demonstrates in general

the same graph as the first if the three lines of growth are compared. Their actual growth over the average period of treatment of 11.7 months was 2.2 inches. Normal increment for their age and length of time corresponding to treatment was only 1.9 inches. Average growth under treatment, then, exceeded the normal increment by three-tenths of an inch. Their assumed growth for this period of time previous to treatment, based upon their actual height and growth rate, as determined from the histories and actual yearly school measurements, was only one-half inch. It is considered, then, that treatment resulted in extra stimulus of growth of 1.7 inches.

An interesting observation is made in patients D.B., J.B. and B.B., who are brothers. The father of these children and one of his sisters displayed a very similar marked type of pituitary dwarfism, tending to bear out the often repeated statement that endocrinopathic states are not infrequently

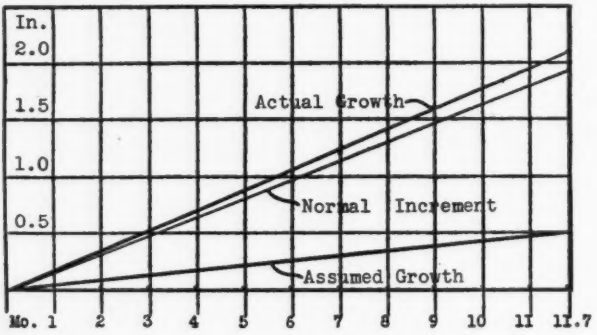


Chart II. Second group. Average duration of treatment, 11.7 mo.; average gain in height on treatment, 2.2 in.; average normal increment for age and time, 1.9 in.; average assumed growth for 11.7 months prior to treatment, 0.5 in.

transmitted from parent to offspring. In our observation of twenty pituitary dwarfs this was the only instance in which such definitely positive history could be obtained. All the offspring of this pituitary dwarf, as presented in figure 1, display signs of marked undergrowth except the eldest. However, she, also, is just below the minimal normal height for her age. In our experience, the family history frequently discloses a tendency on either the maternal or paternal side to short statural height, but not to true nor actual dwarfism.

An achondroplastic dwarf was treated for sixteen months. He gained 3.0 inches in height during this interval. Roentgenographic follow-up studies, together with measurements of the span and upper and lower measurements, suggested that this increase was due to long bone growth rather than torsal bone gowth.

The average human patient does not lend himself well to accurate metabolic nor laboratory studies. We encountered additional difficulties in that a majority of these patients were scattered geographically and not under personal observation. The above studies, therefore, are a compilation of results not from our own laboratory but from several. Only those reports which were

deemed reliable are presented. It must be remembered that these patients were all recruited from families which were able and did attempt to furnish a well-balanced and adequate diet. In no instance was a dietary change suggested. The majority of these patients had the benefit of vitamin and thyroid therapy previous to administration of the pituitary growth hormone.

Blood calcium observations revealed no appreciable change before or during treatment. There was a rise in blood phosphorus, from 3.0 mg. per 100 cc. of blood before treatment to 4.7 mg. during treatment. This alters

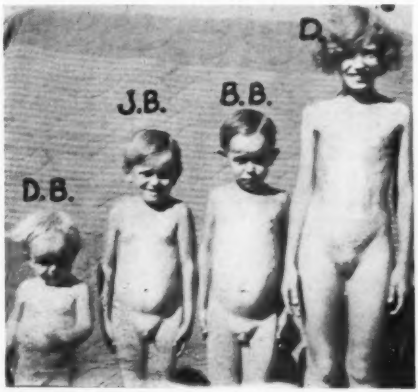


Fig. 1. Patients D.B., J.B., B.B., before treatment. Ages and heights given in table 1, second group. D, sister, untreated; age 10 years, height 50.5 in.

Fig. 2. Father of patients D.B., J.B., B.B., and his complete family. Father x; sister y; 3 brothers and 5 other sisters of normal size. Sister y has 3 normal children.

the Ca-P ratio. There was a relative but definite increase in the blood uric acid determinations, from 2.6 mg. per 100 cc. of blood before treatment to 3.2 mg. during treatment. Blood nonprotein nitrogen remained practically the same before and during treatment. There was a slight but definite decrease in the blood sugar level from 86 mg. per 100 cc. of blood before treatment to 79 mg. during treatment. This same decrease was also noted in the analyses made in our own laboratory. An elevation of the basal metabolic rate, from plus 12 per cent before treatment to plus 37 per cent during treatment, was noted. It would be fallacious to place any stress upon the value of the basal metabolic observations because it is realized that the determination in the lower age group is quite inaccurate with the average clinical apparatus available. However, our personal laboratory procedures did display a definite increase in the metabolic rate during treatment as compared with those taken before institution of treatment. It may be of significance to mention that the majority of these observations were made previous to the time that the extract was bio-assayed for the thyrotropic factor. Our later lots of extract have shown experimentally increased animal potency with presumably greater purification as well as elimination of the other intrinsic pituitary factors. Despite this, its application in the human has shown no enhancement of results, admitting that no conclusions can be drawn because of the short period of time during which the more purifed extract was administered. However, this proposes

TABLE 3. METABOLIC STUDIES

Case	Before Treatment							During Treatment						
	Blood Calcium	Blood Phosphorus	Blood Uric Acid	Blood Non-Protein Nitrogen	Blood Sugar	Basal Metabolism	Specific Dynamic Action	Blood Calcium	Blood Phosphorus	Blood Uric Acid	Blood Non-Protein Nitrogen	Blood Sugar	Basal Metabolism	Specific Dynamic Action
	mg.	mg.	mg.	mg.	mg.	%		mg.	mg.	mg.	mg.	mg.	%	
R.S.	11.0	4.4		28	94	+17		8.1	5.2		36	87	+104	
P.G.	11.3	4.6		29	60	−29	7	8.1	2.9		33	90	+ 11	9
T.W.				30	80						28	84		
J.C.	9.8	4.2		35	100			9.2	5.2		25	78		
D.B.				36	50	+67					29	45	+ 78	
J.B.				28	100	+14					30	45	+ 73	
B.B.				38	83	+18					28	42	+ 53	
J.D.	9.2	4.0	2.6	25	100	+ 6	11	8.4	5.3	2.8	26	100	− 1	12
L.W.	9.8	4.6						9.0	4.7					
M.P.	9.0	3.8	3.4	26	87	+ 2	−9	9.2	4.6	3.4	25	109	+ 8	12
F.O.	10.8	3.2	2.1	26	95	+ 5	42	9.0	5.0	3.5	27	95	− 1	−13
C.I.	9.2							9.8						
J.I.	8.8							10.2						
T.S.	10.6	3.1	2.3	25	92	+ 6	2	9.8	5.0	3.0	21	89	+ 7	2
Average	9.9	3.0	2.6	30	86	+12	11	9.7	4.7	3.2	28	79	+ 37	4.4

the question as to whether an admixture of these various pituitary hormones might not prove to be the therapeutic epitome in human dwarfism. Those cases displaying even markedly elevated basal metabolic rates during treatment did not at any time show thyrotoxic signs.

The specific dynamic action determinations must be placed in the same category as the basal metabolic rates, for the same difficulties are met in this procedure. The depression from 11 before treatment to 4.4 during treatment is contrary to our concept of the purpose for which this test was originally designed, viz., that in pituitary hypofunction it should be low and that in pituitary hyperfunction it should be comparatively increased. One would then be led to believe that the administration of this extract should elevate rather than depress. In our hands it has proven of little value in the diagnosis of pituitary dwarfism or of general endocrine dysfunction.

CONCLUSIONS

The pituitary extract employed was the responsible factor in the production of growth in these dwarfed human beings. Thyroid medication has proven clinically and experimentally a valuable adjunct in treating pituitary dwarfism.

Roentgenologically, a delay in osseous development is a good prognostic sign. Contra-indications for the administration of this extract are epiphyseal closure and approaching sex maturity.

SUMMARY

Eleven cases of pituitary dwarfism of varying degree have been treated from 6 months to 2 years and 9 months with anterior pituitary extracts. Statural height in excess of the normal growth for their individual ages and period of treatment has been induced. Proper selection of cases for this treatment depends upon a comprehensive diagnostic survey to rule out non-endocrine disease. An important diagnostic procedure is mensuration, in that the patient's measurements should be proportionate and all below the minimal normal, bearing in mind that the true pituitary dwarf is a proportionate individual just as is the pituitary giant. Generalized chondro-epiphyseal changes including such conditions as Osgood Schlatter's, Perthes' and Kohler's disease are not an infrequent finding. It has previously been demonstrated that this extract was capable of producing amenorrhea in 2 patients with normal rhythmic menstrual periods, and, in an adult male aged 32 years, of destroying libido entirely during the cause of treatment.

We are indebted to Dr. R. G. Hoskins who kindly suggested the metabolic studies.

REFERENCES

1. Engelbach, W., R. L. Schaefer, and W. L. Brosius: Endocrine growth deficiencies: diagnosis and treatment. Endocrinology, **17**: 250. 1933.
2. Engelbach, W., and R. L. Schaefer: Endocrine dwarfism: Second report. Endocrinology, **18**: 387. 1934.
3. Smith, Philip E.: Increased skeletal effects in A. P. growth-hormone injections by thyroid in hypophysectomized, thyroparathyroidectomized rats. Proc. Soc. Exper. Biol. & Med. **30**: 9. 1933.

THE CLINICAL SIGNIFICANCE OF ELECTRICAL IMPEDANCE DETERMINATION IN THYROID DISORDERS*

J. W. HORTON,[1] A. C. VANRAVENSWAAY,[2] SAUL HERTZ[3] AND G. W. THORN[4]

From the Thyroid Clinic of the Massachusetts General Hospital

BOSTON, MASSACHUSETTS

Interest in the relationship between the phase angle of the electrical impedance of the human body and thyroid disturbances has been prompted by the recent work which Brazier (1), (2), (3), (4) has reported in England. During the past year the American Goiter Society granted Brazier its first award for her contribution to the study of thyroid disorders. Furthermore, at the present time an apparatus of the type described by Brazier is being offered for sale in England as a diagnostic instrument for use in thyroid disease. According to the publication of this author it is possible to follow the clinical course of thyroid disease very accurately, using this electrical measurement as the basis for the estimation of thyroid activity. Since impedance values are little affected by food, activity, menstrual cycle and so forth, it appeared that this measurement might offer decided advantages over metabolic rate determinations.

The earliest attempts to measure the impedance of the human body were made with direct current. Using a direct current, the impedance offered by the body to the current has but a single component, namely, the resistance. However, when an alternating current is employed, the body conducts the current as would a circuit containing both a resistance and a capacitance. In other words it exhibits both resistance and reactance. The ratio of the values of these two components is a property of the tissues under observation. Brazier believes this property to be a function of thyroid activity. The ratio of the reactance to the resistance Brazier defines as the "Impedance Phase Angle." Inasmuch as this ratio is not represented by an angle in conventional electrical engineering practice we shall avoid the use of this expression. Instead we shall follow the notation, already well established, in which this ratio is designated as the factor "Q."

The method of making electrical contact to the body recommended by Brazier involves immersing the arms to the elbows in saline solution. It is claimed that by thus making the area of the surface tissues relatively large their effect upon the impedance measure is negligible. That this is far from true has been demonstrated by Horton and VanRavenswaay, who have developed a method for measuring separately the impedances of the surface sheath and of the internal tissues. The method used by them involves the use of four electrodes which, in effect, form the terminals of a multiple branch electrical network. Certain of the branches being due solely to surface tissues and others solely to internal tissues the evaluation of

---

* Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 11, 1935.

[1] Research Associate, Department of Electrical Engineering, Massachusetts Institute of Technology, Cambridge, Massachusetts.

[2] Resident, Department of Pathology, Massachusetts General Hospital.

[3] Research Fellow, Department of Medicine, Harvard Medical School and Massachusetts General Hospital.

[4] Rockefeller Fellow in Medicine, Harvard Medical School and Massachusetts General Hospital.

each may be effected by computation from measurements of the impedances between the several possible pairs of terminals. There has also been developed an impedance comparator, employing the principle of the alternating current potentiometer, by which it is possible to obtain by direct measurement the values of the

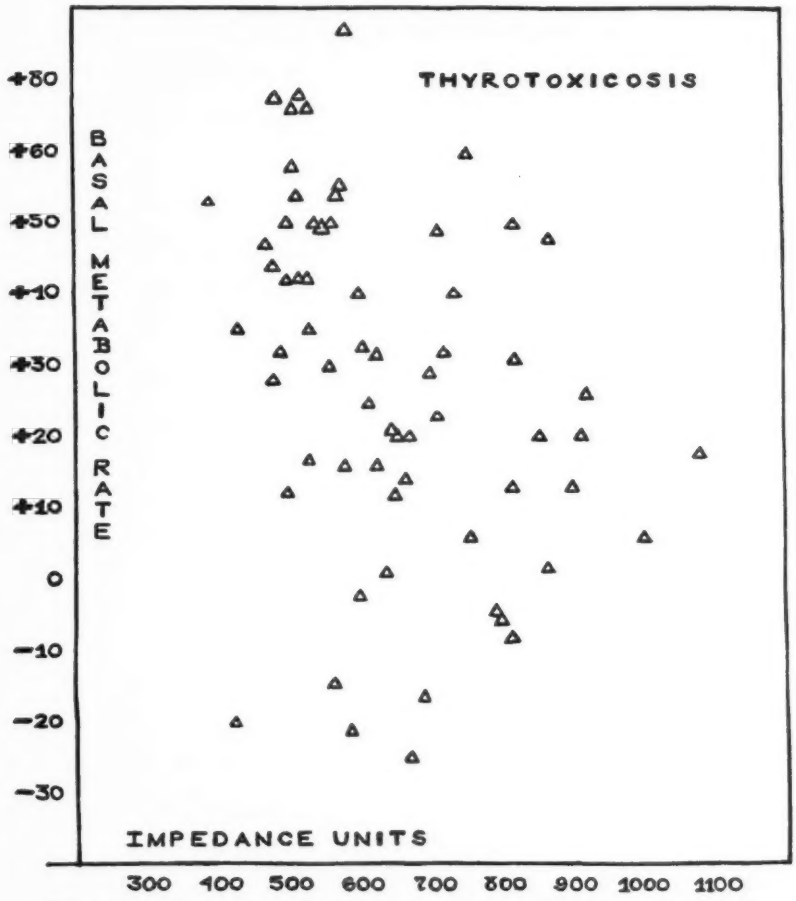


Fig. 1. Impedance values of treated and untreated thyrotoxic patients at different metabolic levels.

resistance and of the reactance components of the surface sheath areas under each of two electrodes and of the internal tissues joining these areas.

Data obtained by the four electrode method, both with the comparator and by the direct measurement on the several electrode pairs, lead to the following conclusions. A detailed discussion of this part of the work is given in another paper (5).

(1) The characteristic curves showing the variations of the impedance components with the frequency for the surface sheath are markedly different from those for the internal tissues.

(2) The value of Q for the surface sheath, at 10 kilocycles, may vary between

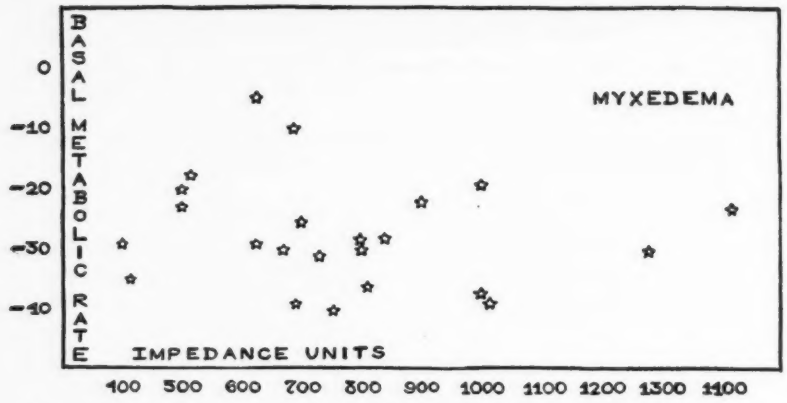


Fig. 2. Impedance values of treated and untreated myxedematous patients at different metabolic levels.

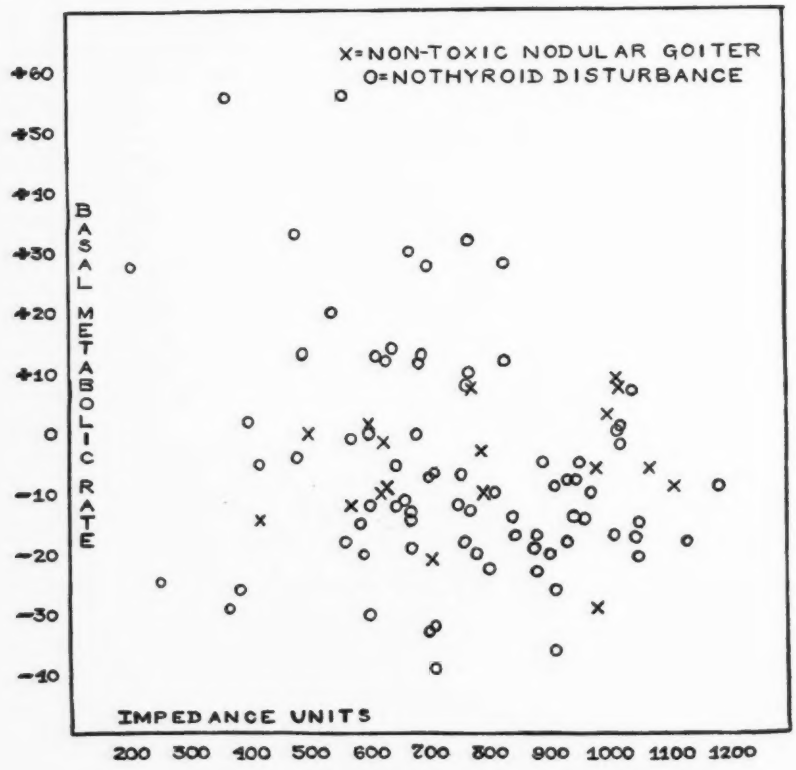


Fig. 3. Impedance values in patients with non-toxic goiter, non-thyroid disorder and no demonstrable disease.

1 and 4 for different individuals. It may vary by a factor of 2 for different positions on the surface of a given individual. It may also vary by a factor of 2, or more, over a period of several days for a given area.

(3) The value of Q for the internal tissues, at 10 kilocycles, may vary between 0.04 and 0.14 for different individuals. It appears to vary but little (less than 10 per cent) with the portion of the body included between the electrodes for a given individual. With persons in normal health the change over long periods of time

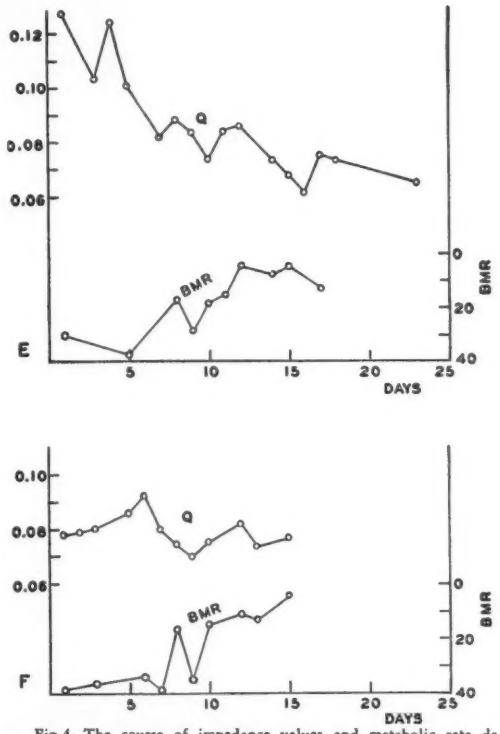


Fig.4. The course of impedance values and metabolic rate determinations in two typical cases of myxedema (E,F) on thyroid therapy.

is, in general, less than 10 per cent. With pronounced changes in pathological condition it may change by as much as a factor of 2.

(4) Using representative values for the impedance components of the internal and surface tissues, per unit length of arm, the total impedance of an immersed arm, between the electrolyte and the internal tissues lying in the plane of the electrolyte surface, has been computed. The results show that of the total impedance measured by the immersion method the portions of the arms below the surface of the electrolytes contribute less than 10 per cent of the resistance and nearly 50 per cent of the reactance. As a consequence the values of Q given by the immersion methods are, in general, from 50 to 100 per cent greater than the values for the internal tissues alone. They are obviously greatly affected by changes in the impedance of the surface sheath.

The conclusions outlined above suggest strongly that any study of possible correlations between electrical impedance and pathological conditions should consider the surface sheath and the internal tissues separately. In the clinical investigation which forms the subject of this report, the electrical constant to which particular attention has been given is the ratio of the reactance to the resistance components or the impedance of the "internal tissues," measured between band electrodes placed on the upper arms.

This report is based upon a study of 220 individuals, representing more

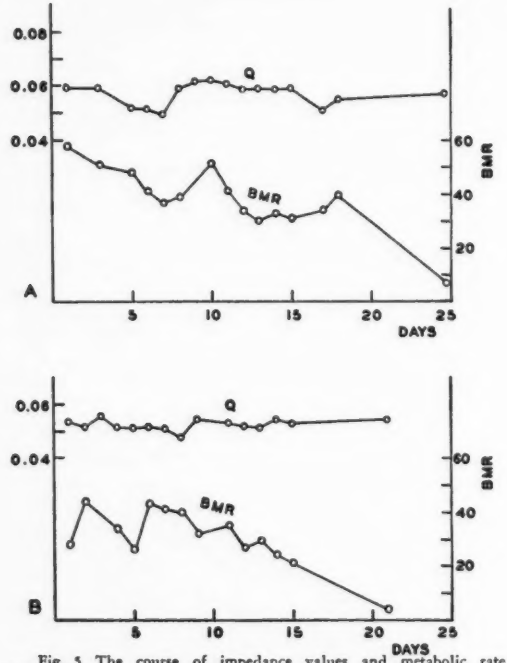


Fig. 5. The course of impedance values and metabolic rate determinations in two typical cases of thyrotoxicosis (A,B) under treatment by iodine and subtotal thyroidectomy.

than 700 impedance records accompanied by simultaneous basal metabolic rate determinations in nearly every instance. The cases were grouped as follows.

Thyrotoxicosis	63
Myxedema	25
Non-Toxic	18
Non-Thyroid Diseases	72
Normal Individuals	42

The plan of study included a general survey of all cases with particular reference to thyroid function as measured by basal metabolic rates and clinical symptomatology. Cases were studied at various stages of therapy in order to ascertain any correlation of impedance values with the metabolic level. This study included observations on thyrotoxic patients at untreated levels, after

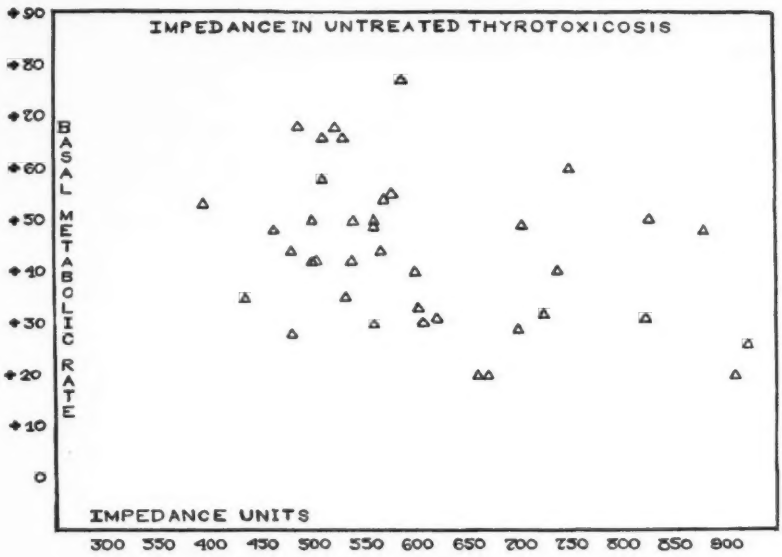


Fig. 6. The impedance values on thirty-nine cases of untreated thyrotoxicosis.

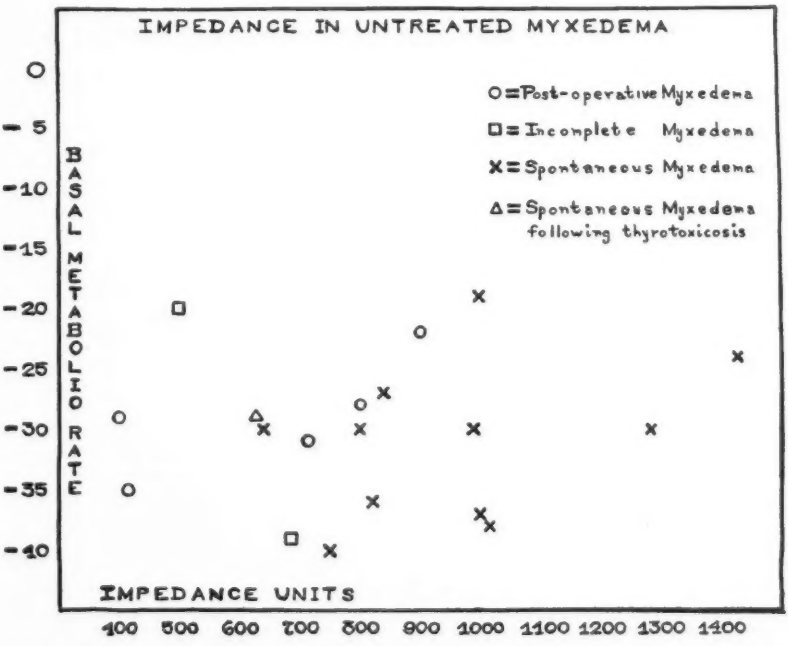


Fig. 7. The impedance values for eleven cases of spontaneous, complete myxedema and eight cases of other types of hypothyroidism.

iodine therapy had been instituted, and at varying intervals following thyroidectomy as well as myxedematous patients before and after the administration of thyroid preparations.

Figure 1 presents data on individuals with thyrotoxicosis at various levels of therapy. Figure 2 presents similar data for myxedematous individuals, while figure 3 gives the values for individuals with non-toxic goiter, with non-thyroid disorders, and those with no demonstrable disease. It is apparent from figures 1, 2 and 3 that there is but little correlation between actual metabolic levels and impedance values.

Cases of thyrotoxicosis and myxedema under therapy showed the usual marked acute changes in their metabolic rates and clinical status. In most instances these alterations were not accompanied by significant changes in impedance values. However, in isolated cases studied over long periods, i.e. six to twelve months, there was a tendency for the high values of myxedematous patients to decrease and for the low values of thyrotoxic patients to increase. Case E in figure 4 indicates a decrease in impedance values in a patient with myxedema on thyroid treatment whereas Case F in figure 4 showed no change in impedance value despite entirely analagous conditions.

Figure 5 shows little change in the impedance values of 2 characteristic cases of thyrotoxicosis (A, B) under iodine treatment and following sub-total thyroidectomy despite marked alterations in their metabolic rate. From these charts, which are representative of the total number of cases followed, it is apparent that great alterations in metabolic rate may occur in the course of treatment of the two groups without regular changes in simultaneously recorded impedance values.

The third part of the study was carried out to ascertain whether the impedance determinations in untreated patients could be used as a diagnostic technic in thyroid disease. Figure 6 illustrates the impedance values obtained on 39 cases of untreated thyrotoxicosis. The range of impedance values for this group was from 0.0400 to 0.0910. As indicated in figure 7 the range of impedance values for untreated, complete, spontaneous myxedema (11 cases) was from 0.0650 to 0.1400. Other hypothyroid cases varied from 0.0400 to 0.0900 (8 cases). One case of spontaneous myxedema following the untreated course of thyrotoxicosis had an impedance value of 0.0620. The variation in impedance values for normal individuals, for individuals ill with nonthyroid disorders, and for cases of non-toxic nodular goiter was from 0.0200 to 0.1175.

Thyrotoxicosis	0.0400-0.0900
Spontaneous, complete myxedema	0.0650-0.1400
Other hypothyroid states	0.0400-0.0900
Non-thyroid diseases	0.0200-0.1200
Normal individuals	0.0420-0.1110

For convenience in charting we have in all cases omitted the decimal point in the calculated values of internal impedance; i.e. 0.0910 equals 910 units.

The charts indicate some tendency for the thyrotoxic patients to have low impedance values whereas the myxedematous patients have an opposite trend toward higher impedance values. Neither of the groups was uniform in its

distribution. This fact coupled with the wide range in values for normal individuals and non-thyroid patients vitiates the use of the impedance determination as a diagnostic technic in thyroid disorders.

DISCUSSION

It is important to re-emphasize the fact that the technic used in this study permitted the independent determination of the *internal impedance*. Previous reports on impedance have been based upon a technic in which the contributing components (surface sheath and internal impedance) have not been separated.

Recent data published by Robertson and Wilson, (6), using the Brazier technic failed to confirm many of Brazier's conclusions in regard to the diagnostic significance of impedance determinations in thyrotoxicosis and myxedema. Our own observations made with this method resulted in similar conclusions.

Experimental studies which we have made on animals as well as patients have indicated a remarkable constancy of the impedance value. Studies designed to alter impedance have further emphasized this constancy. Factors such as time of day, ingestion of food, smoking, mild exercise, and prolonged rest in human subjects have produced little or no alteration in the impedance value. Experiments upon cats in which the blood supply to the region studied was obliterated, the nerve injured, NaCl and $CaCl_2$ infused, different layers of the tissue dissected, resulted in very little alteration of the impedance values. Such constancy points toward the existence of compensatory mechanisms in tissues which preclude marked variations in impedance.

The marked variation in impedance values among normal individuals, however, would seem to indicate that there are factors operating in the human body which may alter this electrical quantity. Body weight and surface area seem unimportant in determining impedance values. Up to the present we have been unable to identify any such factors with the possible exception of the level of thyroid activity. We suspect the possible effect of thyroid activity upon the impedance value because of the tendency for the thyrotoxic and myxedematous patients to have widely differing values. This fact may be of academic interest and of bio-physical significance, but certainly it is of little value in aiding the clinical diagnosis of thyroid disease.

SUMMARY

A satisfactory technic has been evolved by Horton and VanRavenswaay for the determination of the internal impedance of the human body as separated from the impedance of the surface sheath.

The internal impedance for any normal individual remains remarkably constant from day to day. Internal impedance varies but little after the ingestion of food, after muscular exercise, and with other factors which alter the metabolic rate.

Using the Horton-VanRavenswaay technic, untreated thyrotoxic patients show a wide range of impedance values (0.0400 to 0.0999). A large percentage of the untreated thyrotoxic patients have low impedance values.

The impedance values in untreated spontaneous myxedema cases range from 0.0650 to 0.1400. The majority of these cases have high values.

Marked alterations in metabolic rates in thyrotoxic and myxedematous patients under treatment are not regularly associated with changes in impedance.

A comparative study of basal metabolic rate and impedance determinations indicate that the latter is of little clinical significance in the estimation of the level of thyroid activity.

REFERENCES

1. Brazier, M. A. B.: J. Instit. Elect. Engineering **73**: 203. 1933.
2. Brazier, M. A. B.: Lancet **2**: 742. 1933.
3. Brazier, M. A. B.: Lancet **2**: 869. 1934.
4. Brazier, M. A. B., and F. M. Grant: Lancet **1**: 125. 1934.
5. Horton, J. W., and A. C. VanRavenswaay: J. Franklin Institute, **220**: 557. 1935.
6. Robertson, J. D., and A. T. Wilson: Lancet **2**: 1158. 1934.

FUNDAMENTAL SIMILARITY IN THE DEVELOPMENT OF GONADOTROPIC RESPONSE IN THE IMMATURE GUINEA PIG AND RAT

S. C. FREED AND A. COPPOCK

From the Department of Metabolism and Endocrinology and the Department of Pathology, Michael Reese Hospital

CHICAGO, ILLINOIS

The difference in response to the administration of human pregnancy urine (P.U.) between the guinea pig and the rat, has been described by several workers. According to Loeb (1) the changes observed in the guinea pig ovary are chiefly those of thecal luteinization, with some maturation of the granulosal cells. King (2) and Selye et al. (3), also described this thecal luteinization, and the latter authors compared their results to the effects obtained in the hypophysectomized rat. On the basis of this similarity they suggested that the failure of the guinea pig to give the typical Aschheim-Zondek reaction was due to a lack of secretion of the complementary substance from this animal's hypophysis.

The refractoriness of the immature rat to P.U. up to the age of about 18 days, is well-known (Selye et al., 4). P.U. extracts are similarly ineffective in infantile rabbits up to about 12 weeks of age (Hertz and Hisaw, 5). It is only after these respective periods that the gonadotropic hormones become appreciably effective. In the present work, a study of the effects of P.U. extracts on the guinea pig and rat at various ages revealed the fact that the former animal need not be regarded as differing materially in its response from the normal rat (or rabbit). The apparent differences previously reported are in reality quantitative rather than qualitative, and depend upon the fact that isolated periods rather than the whole cycle of events have heretofore been studied.

METHOD AND RESULTS

Immature guinea pigs, at varying ages, were injected with large doses of P.U. extract, prepared by the Katzman and Doisy (6) benzoic acid method. Daily doses, varying in different animals from 100-400 rat units, were administered for four successive days, the animals being killed on the sixth day. The procedure on rats was similar except that a daily dose of 25 rat units of P.U. extract was used.

Because we did not breed the guinea pigs we used, we cannot vouch for their exact ages. It is sufficient for our present purposes, however, to classify our results according to the weight and approximate age of these animals. For purposes of comparison our results on both species are given in parallel columns, in the following table.

Guinea Pig	*Rat*
Dose—100 — 400 R.U. × 4	*Dose*—25 R.U. × 4
Stage I	
Age—0 to 2 weeks (110 gm. or less)	[1]*Age*—0 to 6 days
Ovary—Negative (some follicular atresia)	*Ovary*—Negative
Uterus—Negative	*Uterus*—Negative
Stage II	
Age—2 to 3-4 weeks (110-140 gm.)	[1]*Age*—6 to 15-16 days
Ovary—Thecal luteinization of those follicles which bear antra. Slight thecal hypertrophy about follicles without antra	*Ovary*—Thecal luteinization of follicles, with or without antra
Granulosal cells not affected	Granulosal cells not affected
Uterus—Negative	*Uterus*—Estrous hypertrophy varying with age; uterine weights:—
	Age / Before injection (average) / After injection (average) 8 days / 6 mg. / 20 mg. 12 days / 10 mg. / 50 mg. 15 days / 12 mg. / 70 mg.
Stage III	
Age—3-4 to 8-9 weeks (140-220 gm.)	[1]*Age*—15-16 to 16-17 days
Ovary—	*Ovary*—
a) *Early phase* (140-160 gm.) Thecal luteinization of follicles, with or without antra Granulosal cells of larger follicles show increased cytoplasm and lighter staining nuclei	
b) *Late phase* (160-220 gm.) Heavier thecal luteinization Granulosal cells of small follicles with or without antra, show changes described in (a) In larger follicles these cells approach size and appearance of lutein cells, but no capillary invasion	Heavy thecal luteinization Some granulosal cells show transitional changes as described in guinea pig, but rapidity of change is such that complete luteinization is already seen in other portions of ovary, but true corpora not yet formed.
Uterus—Slight hypertrophy and hyperemia	*Uterus*—Estrous hypertrophy
Stage IV	
Age—8-9 weeks to maturity (12-13 wks.)	[1]*Age*—18 days to maturity
Ovary—	*Ovary*—(Typical Aschheim-Zondek reaction)
A few large corpora lutea with complete granulosal luteinization, and abundant capillary invasion.	Several large corpora lutea with complete granulosal luteinization, and abundant capillary invasion
Occasional "lutein cyst"	Frequent "lutein cysts"
[2]Remaining follicles as in III (b)	
Uterus—Moderate hypertrophy	*Uterus*—Estrus
Progestational proliferation (Cuboidal epithelium)	(Columnar epithelium)

[1] On day of first injection.
[2] The heavier thecal luteinization in the guinea pig as compared to the rat may, on superficial examination, obscure the fundamental similarity of the remaining structures. The corpora in the guinea pig ovary also tend to be less hemorrhagic than those in the rat.

DISCUSSION

It may be seen that the ovarian effects obtained with P.U. in the guinea pig are essentially similar to those obtained in the rat, except that the time intervals involved are different. Part of this difference might be interpreted on the same basis that Hertz and Hisaw explained the development of responsiveness in the ovaries of their immature rabbits. According to these workers, only those follicles bearing antra are susceptible to hormonal influence. Our results are in accord with this view insofar as the larger follicles underwent the

most striking changes. It must be pointed out, however, that with increasing maturity of our guinea pigs, the entire ovary, including the smallest follicles increased in sensitivity. Some of the difference between the two species may, therefore, depend upon a difference in the rate of change in those properties of the ovary which, for lack of more precise knowledge, have been called the intrinsic nature of the ovarian cells.

As regards the difference in response of the uterus of these animals, the

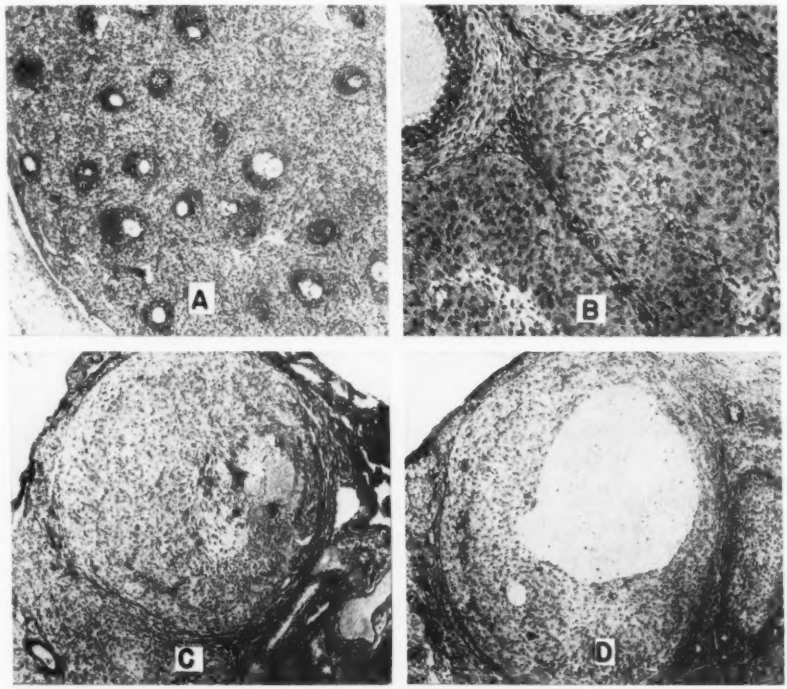


Fig. 1.

A. Ovary of treated guinea pig (130 gm.) Stage II, thecal luteinization only. ×80.
B. Ovary of treated guinea pig (190 gm.) Stage III, heavy thecal luteinization. Granulosal cells in transitional stage. ×175.
C. Ovary of treated guinea pig (220 gm.) Stage IV, Corpus luteum, capillaries invading granulosal lutein cells. Also small follicle with granulosal cells in transitional phase. ×80.
D. Ovary of treated guinea pig (216 gm.) Stage IV, "Lutein cyst". ×80.

lack of the estrus phenomenon in the guinea pig is not as great a dissimilarity as might seem on first thought. Since it is well known that the presence of estrin is necessary to prepare the uterus for the progestational proliferation caused by the lutein hormone, the guinea pig ovary must have been stimulated to produce the necessary amount of estrin. The fact that this amount of estrin was apparently insufficient to produce typical estrus, is again a quantitative and not a qualitative difference.

Whatever be the cause of the difference between the responses of the guinea pig and rat, a comparison of the length of the various stages in these

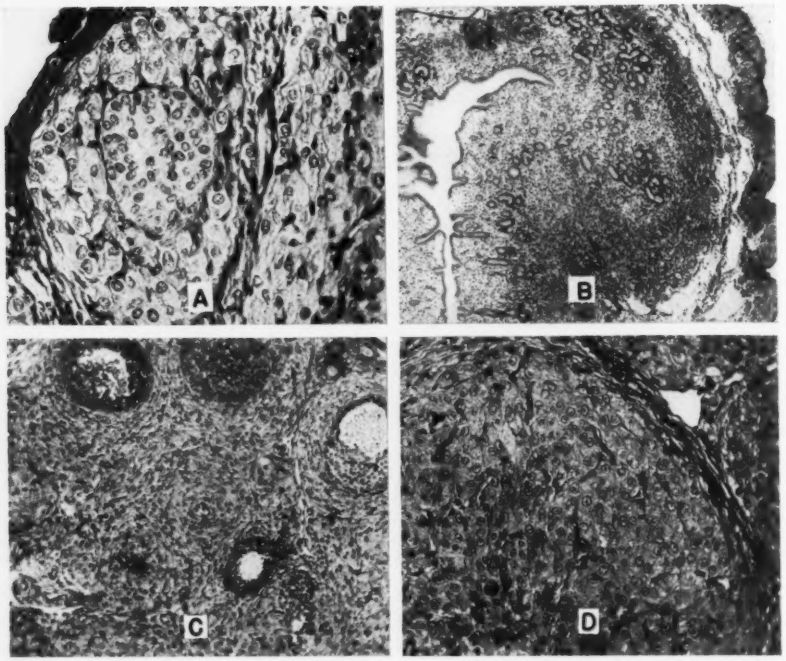


Fig. 2.

A. Same ovary as Fig. 1.D., showing transitional cells of granulosal origin in primary follicle. ×325.
B. Uterus of animal whose ovaries are shown in Figs. 1.D. and 2.A., showing progestational proliferation. ×175.
C. Ovary of rat (15 days) Stage III, several follicles showing, simultaneously, all gradations between normal granulosal cells and ripe lutein cells. ×175.
D. Another field of ovary shown in Fig. 2.C., showing granulosal luteinization in a lutein body. ×325.

animals, as shown in figure 3, shows that the greater portion of the immature life of the guinea pig is spent in stages two and three, during which there is relatively little response to P.U. The fact that these stages coincide with the size and age of the guinea pigs usually employed for hormone testing, probably accounts for the supposed peculiarity in their reactions. Our work therefore disposes of an example of those species differences, which have been a confusing element in our newer knowledge of the hormones.

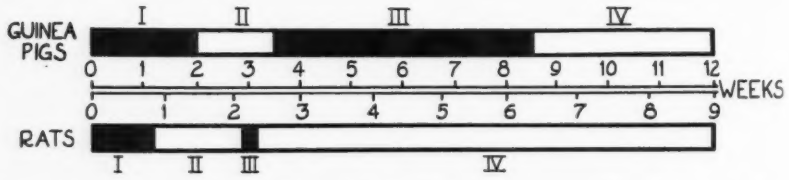


Fig. 3. Graphic comparison of the periods of sexual immaturity in the guinea pig and rat; showing the proportionate span of the various stages of immaturity which have been described for these animals respectively. Note the relatively great length of Stage III in the guinea pig as compared to the rat.

SUMMARY

The response of the guinea pig to the administration of pregnancy urine is essentially similar to that of the rat. Some of the factors responsible for the apparent difference between these species are demonstrated and discussed.

We wish to express our appreciation for the aid and direction of Dr. Samuel Soskin.

REFERENCES

1. Loeb, L.: Endocrinology **16:** 129. 1932.
2. King, A. G.: Proc. Soc. Exper. Biol. & Med. **30:** 1182. 1933.
3. Selye, H., J. B. Collip, and D. L. Thompson: Proc. Soc. Exper. Biol. & Med. **30:** 780. 1933.
4. Selye, H., J. B. Collip, and D. L. Thompson: Proc. Soc. Exper. Biol. & Med. **32:** 800. 1935.
5. Hertz, R., and F. L. Hisaw: Am. J. Physiol. **108:** 1. 1934.
6. Katzman, P. A., and E. A. Doisy: J. Biol. Chem. **106:** 125. 1934.

CHRONIC HYPOGLYCEMIA[1]

MAX A. GOLDZIEHER
NEW YORK CITY

The incidence of abnormally low blood sugar with or without clinical manifestations has been frequently observed since chemical analysis of the blood has become one of the ordinary diagnostic procedures. Major interest has been focused upon this question since Harris (1), Wilder (2) and others demonstrated that pathological island tissue is capable of producing an excess of insulin and concomitant symptoms comparable to those of insulin shock. Low fasting blood sugar readings were obtained, however, also in cases which presented a variety of symptoms not necessarily associated with the incidental overdosage of insulin. The peculiar and varied symptomatology of these hypoglycemic states has been the subject of several comprehensive papers in the past few years and does not need to be elaborated upon. Suffice to refer to the monographs of Sigwald (3) in the French and Wilder (4) in the German literature, as well as to the papers of Tedstrom (5), Wauchope (6) and Cammidge (7). The symptoms of hypoglycemia relate to the psyche, behaviour or consciousness of the patient and they may be accompanied by disturbances of speech, muscular twitchings or convulsions of epileptiform character. Excessive hunger, abdominal pain or nausea and vomiting are concomitant symptoms on the part of the gastro-intestinal tract.

The cause of hypoglycemia appears to be established in those cases in which surgical inspection or autopsy reveals the presence of an adenoma or adenocarcinoma of the pancreatic island tissue. The interpretation of a low blood sugar as the expression of hyperinsulinism is also acceptable, if microscopic examination of the pancreas presents abnormally large or too numerous islands. For the other cases without demonstrable pancreatic pathology the concept of a functional disturbance has been advanced in the sense of a relative hyperinsulinism, that is preponderance of island tissue secretion over the joint performance of its physiological antagonists. Harris, protagonist of this viewpoint expressly distinguishes between hyperinsulinism as the result of excessive island tissue (adenoma, or carcinoma of the islands) and relative hyperinsulinism which might be divided according to the secondary diagnosis into adrenal, pituitary, thyroid and hepatic types.

A recent survey of Tedstrom, analysing 68 cases of hypoglycemia, lists 53 as cases of hyperinsulinism. The hypoglycemia in the remaining 15 instances was explained as the result of liver damage, with the exception of two cases in which extrapancreatic endocrine factors were accepted; one case is listed as carcinoma of the adrenals, and the other as "pluriglandular disease." Among the 53 cases of hyperinsulinism 38 were labeled "functional" while organic lesions of the pancreas were demonstrable only in 15 patients.

The concept of functional hyperinsulinism leads with necessity to the surgical approach of the problem and grossly normal pancreatic tissue has

[1] Read before the Nineteenth Annual Meeting of the Association for the Study of Internal Secretions, Atlantic City, June 10, 1935.

been resected already in a number of cases in order to relieve the symptoms of hypoglycemia. However, the results published by Finney (8), Graham and Womack (9) and others are far from satisfactory. Even substantial resection of the pancreas left the patients unimproved and more favorable reports are incidental and not convincing.

It would thus seem that the concept of hyperinsulinism does not do justice to all cases of hypoglycemia and may provoke unsatisfactory and undesirable surgery. Moreover, study of patients whose blood sugar is low and who may or may not present some of the symptoms known to accompany hypoglycemia, reveals weighty evidence that points to other glands and not to the pancreas as the source of the metabolic disturbance.

In the course of the systematic endocrine investigation of about 2000 patients, we observed 112 cases in which either the fasting blood sugar was found to be abnormally low or a sugar tolerance test revealed hypoglycemic tendencies (fig. 1). The fasting blood sugar readings of these patients varied

TABLE 1.

	Total	Pituitary	Hypo-Thyroid	Gonadal	Improved
Abdominal pain	22	11	11	—	15
Obesity, craving for carbohydrates	18	18	—	—	11
Obesity, excessive appetite	16	13	—	3	9
Dizziness, mild fainting spells	17	14	3	—	12
Tachycardia	16	11	4	1	10
Typical hypoglycemic attacks	8	8	—	—	5
Epilepsy	8	8	—	—	6
Dysmenorrhea	4	3	1	—	3
Speech defects	3	2	1	—	3
Total	112	88	20	4	74

from 53 to 111 with an average of 80 mg. per cent. After ingestion of 0.8 grams of glucose per pound of body weight an increase of the blood sugar was obtained the peak of which varied from 85 to 195 with an average of 111.5 mg. per cent. The drop of the blood sugar after 3 hours varied from 50 to 100 with an average of 72.5 mg. per cent and at 4 hours a further drop was noted, varying from 45 to 80 with 64 mg. per cent as the average figure. The lack of uniformity in sugar tolerance among these hypoglycemic patients is better visualized if we classify the curves obtained according to the relation of fasting level, peak and endpoint. See figure 3. It seems that six different types can be distinguished: a) low fasting level, normal rise and return to the fasting level; b) low fasting level, small rise and return to the fasting level; c) normal fasting value, no rise and slow, consistent drop below the fasting level; d) low normal fasting value, small rise and substantial terminal drop; e) low fasting level, sudden high rise, slow return and deep terminal drop (diabetic type); f) very low fasting level, fairly good rise and early drop to a low point. All these types are found with equal frequency except type c) which is twice as common as any of the other types.

The 112 cases are classified in table 1 according to the chief complaint

of the patients and the endocrine diagnosis made. The age of these patients varied from 8 to 57 years; 93 were females and 19 males. The hypogonadal group includes 3 women with menopausal symptoms and 1 eunuchoid male. The coincidence of hypopituitarism seemed probable in these patients, but could not be definitely diagnosed. The hypothyroid group was characterized by dry skin and hair, low basal metabolic rate and often by shortness of the lower half of the body. The hypopituitary group is the largest and includes 1 case of Simmonds' disease, 1 diagnosed as basophile adenoma, 14 cases of adiposo-genital dystrophy, while the others were characterized by the typical fat distribution, postural hypotension, inadequate rise of the metabolic rate after a protein meal, typical blood picture and dysplasia of the cranium on x-ray examination.

In 1 case of the hypopituitary series, seen only in consultation, the pos-

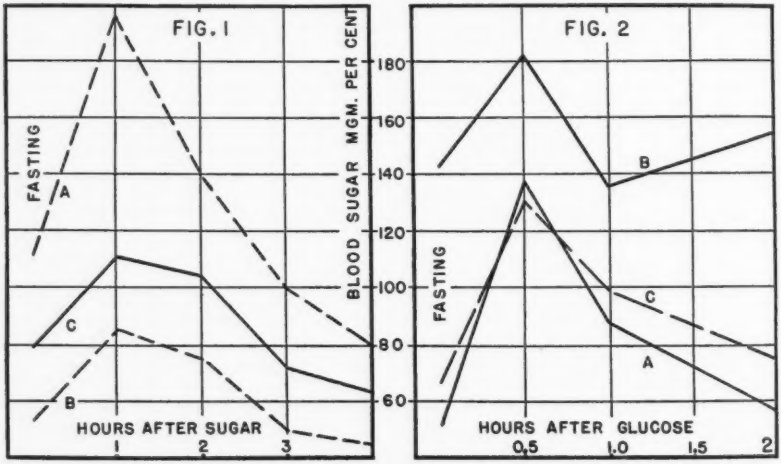


Fig. 1. A and B, extreme curves of sugar tolerance values and C, average of values found in 112 patients after ingestion of 0.8 grams of glucose per pound of body weight.
Fig. 2. Sugar tolerance test in a patient: A, before partial pancreatectomy; B, two days after operation; C, one week after operation.

sibility of hyperinsulinism had been considered. The patient displayed symptoms which were readily explained by attacks of hypoglycemia and the diagnosis was confirmed by determination of a low fasting blood sugar. A sugar tolerance test (fig. 2) showed a rise in one-half hour from a fasting level of 52 to 136, a drop to 88 in 1 hour and a 2 hour reading of 57 mg. per cent. The patient was operated on by Dr. H. Koster of Brooklyn at the Crown Heights Hospital and what appeared to be a normal pancreas was seen and palpated. Nevertheless resection of about two-thirds of the pancreatic tissue was decided upon. On histological examination of the pancreas nothing unusual could be seen. The Langerhans islands were perfectly normal, both as to number and size. A sugar tolerance test made two days after the operation gave cause for hopes. The figures indicated a decreased sugar tolerance of almost diabetic type. The effects of the pancreatic resection, however, wore

off one week after the operation and a sugar tolerance curve obtained at that time was of the hypoglycemic "a" type. See figure 2. The sugar tolerance remained unchanged thereafter as shown by repeated tests in the course of the following few months. Thus the effect of the operation was ephemeral both upon the fasting blood sugar and sugar tolerance. Nor did the symptoms of the patient improve; they were controlled, however, to a tolerable extent by dietetic measures.

This single observation is well in accord with other reports on the failure of pancreatic resection to relieve hypoglycemia or its symptoms in the absence

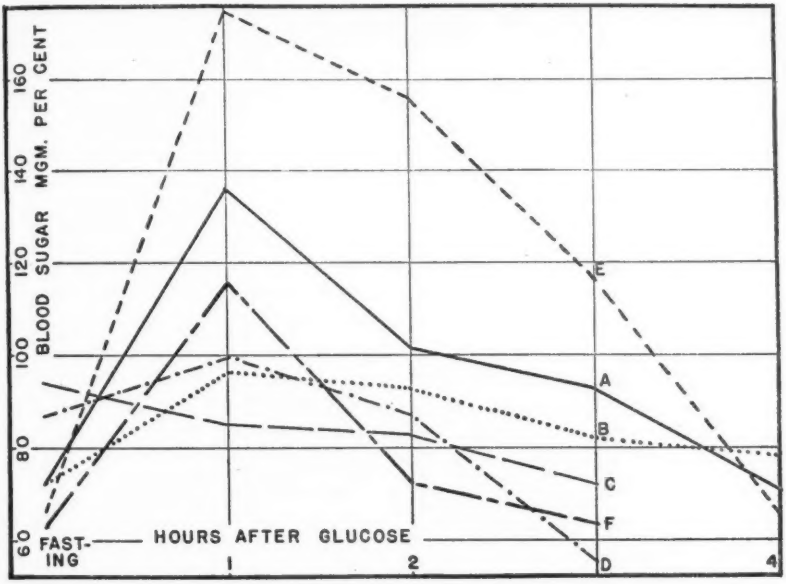


Fig. 3. Types of individual sugar tolerance curves found.

of a tumor of the pancreas. The futility of pancreatic surgery is the more striking, if we compare it with the therapeutic results presented in this paper. Seventy-four cases are listed in the chart as improved which means that these patients were free of their original complaints for some time prior to and at the time of their discharge. Exacerbation of symptoms during treatment was not observed in any case. Several patients whom we could follow for a period of 1 to 3 years after their discharge have remained free of symptoms. The unimproved 38 cases include patients who did not return for treatment or who discontinued it after a short while.

Our treatment consisted of organotherapeutic and dietary measures. The latter varied according to the nutritional state of the patient. Stout patients were given a 900 calorie 40 per cent protein diet with very little fat, the bulk being made up of 5 and 10 per cent carbohydrates. Underweight patients were given a diet of 20 per cent above their calculated caloric require-

ments, rich in proteins and poor in fat. The 5 and 10 per cent carbohydrates were distributed over at least 6 feedings daily. Meals containing substantial amounts of carbohydrates were carefully avoided, as they seemed to precipitate hypoglycemic reactions 3 to 4 hours after ingestion. The excessive appetite of the obese patients was successfully curbed by administration of ephedrin sulfate 3/8 grain orally 2 to 2½ hours after each meal. Administration of ephedrin could be discontinued as a rule after a short while and the drug had to be used thereafter only incidentally.

The hypothyroid patients received desiccated thyroid gland orally; the dosage varied from ¼ to 4 grains daily according to individual requirements. The lean patients with tachycardia, as a rule, needed only small doses and were intolerant to more massive medication. Disappearance of the abdominal pain was the most conspicuous reaction to the treatment. Some of these patients had been diagnosed as cases of chronic appendicitis or possible duodenal ulcer. Two patients had been operated on and had their normal appendices removed without improvement. They obtained relief, however, in a remarkably short time by thyroid medication. Similar observations have been recorded in the older literature, yet they did not attract much attention. A recent paper by Watkins (10) emphasizes again the extent of apparently unnecessary surgery to which these patients are so frequently subjected.

In the pituitary group the dietary measures were combined with injections of a fresh anterior lobe extract. Normalization of the irregular menstrual periods or rapid development of retarded testicular growth were the objective signs by which the efficacy of the organotherapy was gauged. The hypoglycemic symptoms disappeared gradually within a few weeks during the course of treatment. We considered clinical improvement as indication of an adjustment in carbohydrate metabolism. Control blood sugar examinations and sugar tolerance tests after clinically successful treatment were available only in a limited number of cases. They seemed to corroborate the clinical findings although the tolerance curves retained a tendency to flatness.

These therapeutic results are presented to show that patients complaining of hypoglycemic symptoms are likely to respond favorably to treatment that is based on the correct diagnosis of the prevailing endocrine disturbance. This discussion is meant to emphasize first of all the frequency of chronic hypoglycemic states and their alimentary exacerbations; thereafter, it stresses the outstanding rôle of the pituitary and thyroid glands in the pathogenesis of this condition. Deficiency of the adrenals as well as impairment of the liver are readily admitted as other possible causes of the hypoglycemic syndrome, yet these conditions are met with only incidentally and rank in importance far behind pituitary or thyroid insufficiency.

Hyperproduction of insulin is, in our opinion, only one of the rare causes of the hypoglycemic symptom complex. The failure of surgical intervention in the absence of tumors of the pancreas supports this contention. The incidence of island tissue tumors, either benign or malignant, is definitely rare in my own extensive autopsy experience which is in accord with the data available from the literature. These facts, together with the clinical observations reported in this paper, indicate that hyperinsulinism is only rela-

tive in the majority of the hypoglycemic conditions the organic background of which should be looked for in the pituitary or thyroid, and not in the pancreas.

The rôle of thyroid insufficiency in the production of symptoms comparable to those observed in hypoglycemia, has been extensively discussed in the monographs of Hertoghe (11) in 1899 and of Levi (12) in 1922. Wagner and Parnass (13) report on their therapeutic results after thyroid medication and more recently La Roche-Boissiere (14) refers to the significance of thyroid failure in the pathogenesis of the hypoglycemic syndrome. While other French authors emphasize the adrenal angle of the problem, and report beneficial results of adrenal medication (Gougerot and Peyne, 15), Wilder stresses the pituitary factor in the hypoglycemic syndrome. Although he admits the varied origin of the cases described by others or observed personally, he points out the fact that most of the patients were of the hypopituitary type and general appearance.

The significance of pituitary insufficiency in the production of hypoglycemia cannot be questioned after the contributions of Houssay (16), Lucke (17), Collip (18), Braier (19), and others who showed that the blood sugar of the hypophysectomised animal is either low or prone to drop excessively on prolonged fasting. Simultaneously the animals acquire abnormal sensitivity to insulin (Houssay, Lucke). A decrease of liver glycogen has also been noted (Nakamura, 20, Corkill, 21), as well as an inadequate response to the injection of adrenalin, (Aschner, 22, Braier). If it is permissible to draw any analogy from animal experiments to human pathology, it is fair to assume that functional deficiency of the anterior lobe of the hypophysis affects carbohydrate metabolism in the sense of lowering the fasting blood sugar, increasing insulin sensitivity and decreasing the response to other endocrine factors (adrenal, thyroid) which constitute important links in the chain of endocrine regulations.

The evidence of thyroid influence upon the blood sugar level is less clearly established but still convincing. Higher blood sugar readings and decreased sugar tolerance have been noted in hyperthyroidism (Offenbacher and Hahn, 23, Chiba, 24), and the opposite has been shown after thyroidectomy (Eppinger, Falta and Rudinger, 25). It is also known that administration of thyroid hormone mobilizes glycogen from its depots, especially from the liver, hence the frequency of liver damage in hyperthyroidism. It is no contradiction that the liver glycogen is also low in the thyroidectomised animal and in myxedema, partly because of liver damage and partly because of inadequate resorption and utilization of the ingested carbohydrates in hypothyroidism (Parhon, 26).

SUMMARY

A series of 112 cases is presented in which the colorful symptoms associated with chronic hypoglycemia were observed. The fasting blood sugar of these patients varied from 53 to 111 with an average of 80 mg. per cent. Sugar tolerance tests showed curves with the peak at an average of 111.5

with variations ranging from 85 to 195 mg. per cent. After 3 and 4 hours the average of the curves dropped to 72.5 and 64 respectively.

The lack of uniformity of the sugar tolerance curves in hypoglycemia makes their classification difficult. Comparing the relations of fasting level, peak of the curve and endpoint at the third or fourth hour, 6 different types can be distinguished.

The most common clinical manifestations of chronic hypoglycemia are: abdominal pain, excessive hunger or craving for carbohydrates with corresponding obesity, mild or severe fainting spells and tachycardia. Epileptiform seizures, speech defects and dysmenorrhea have been also observed.

Clinical classification of the 112 cases showed hypopituitarism in 88 cases while 20 patients presented evidence of hypothyroidism. Seventy-four patients were improved by organotherapy and dietary measures. Resection of the pancreas was undertaken in one case and completely failed to check the symptoms or to influence the hypoglycemia for more than an initial postoperative period.

The significance of pituitary and thyroid insufficiency in the pathogenesis of chronic hypoglycemia is emphasized.

REFERENCES

1. Harris: J.A.M.A. **83:** 729. 1924.
——————: Ann. Int. Med. **7:** 1084. 1934.
2. Wilder et al.: J.A.M.A. **89:** 348. 1927.
3. Sigwald: L'hypoglycemie. Paris, 1932.
4. Wilder: Med. Klin. **26:** 616. 1930.
——————: Deutsch. Ztschr. f. Neurvenh. **112:** 192. 1930.
5. Tedstrom: Ann. Int. Med. **7:** 1013. 1934.
6. Wauchope: Quart. J. Med. **2:** 117. 1933.
7. Cammidge: Brit. M. J. **1:** 818. 1930.
8. Finney and Finney: Ann. Surg. **88:** 584. 1928.
9. Graham and Womack: Surg., Gynec. & Abst. **41:** 728. 1933.
10. Watkins: Ann. Int. Med. **7:** 1534. 1934.
11. Hertoghe: De l'hypothyoidie benigne chronique. Nouv. Icon. Salpêtr. 1899.
12. Levi: Opotherapie endocrin. Livre de France, 1922.
13. Wagner and Parnass: Ztschr. f. d. ges. exper. Med. **23:** 361. 1921.
14. La Roche-Boissiere: Rev. méd.-chir. d. mal. du foie **3:** 481. 1928.
15. Gougerot and Peyne: Bull. soc. Franç. de dermat. et syph. **33:** 12. 1926.
16. Houssay: Rev. franç. d'endocrinol. **9:** 423. 1931.
17. Lucke: Ztschr. f. d. ges. exper. Med. **88:** 65. 1933.
18. Collip: J. A. M. A. **104:** 827. 1935.
19. Braier: Compt. rend. soc. de biol. **107:** 1195. 1931.
20. Nakamura: Fukuoka-Ikwadaigaku-Zosshi, **24:** 18. 1931.
21. Corkill et al.: J. Physiol. **80:** 193. 1933.
22. Aschner: Arch. f.d. ges. Physiol. **146:** 1. 1912.
23. Offenbacher and Hahn: Arch. f. Verdauungskr. **30:** 1. 1922.
24. Chiba: Tohoku J. Exper. Med. **23:** 474. 1934.
25. Eppinger, Falta and Rudinger: Ztschr. f. klin. Med. **66:** 1. 1908.
26. Parhon: Compt. rend. soc. de biol. **83:** 6. 1920.

THE INFLUENCE OF CORTICO-ADRENAL EXTRACT ON THE COURSE OF BACTERIAL INTOXICATIONS IN GUINEA PIGS[1]

RICHARD W. WHITEHEAD AND CHARLES A. FOX
Department of Physiology and Pharmacology, University of Colorado School of Medicine
DENVER, COLORADO

That the secretions of the adrenal cortex play a significant rôle in the resistance of the body to infectious processes seems fairly well established. Morphological findings, both in animal and human material have shown that the adrenal gland, particularly the cortex, is severely injured in acute fulminating intoxications. In chronic infections, an hypertrophy of the cortex is frequently observed. Experimentally, the investigations of Lewis (1), Marine (2), Scott (3), and others have shown that adrenalectomy results in a marked decrease in the natural resistance of animals to bacterial toxins, drugs, and infections. Perla and Marmorston-Gottesman (4) demonstrated that the ability of adrenalectomized rats to produce antibodies is markedly diminished. The administration of potent extracts of the cortex to adrenalectomized animals has been found by Scott and Bradford (5) and Perla and Marmorston-Gottesman (6) to raise the resistance of the animals to within normal values.

In 1933, Scott, Bradford, Hartman, and McCoy (7) studied the effect of extracts of the adrenal cortex on the resistance of unoperated guinea pigs, rats, and mice to different types of intoxications. These workers used diphtheria toxin in the guinea pig, trypanosome infection in the rat, and pneumococcus infection in the mouse. Their results failed to show that any protective influence was afforded by cortical extracts. In the same year, Wenner (8) studied the effect of cortical extract in experimentally produced maxillary sinusitis in rabbits. The experimental animals were treated with an extract of the adrenal cortex and at autopsy the sinuses of the experimental rabbits appeared normal, whereas the controls showed marked inflammation of the sinuses.

A limited number of reports have appeared in the literature regarding the clinical application of cortical extracts in infectious processes. Whitehead and Smith (9) reported a series of 5 cases of acute intoxications, treated with cortical extracts, with beneficial results. Wenner and Cone (10) administered an adrenal extract to patients with pyogenic infections, mostly of the upper respiratory tract, with noteworthy effects.

The findings of Scott, Bradford, Hartman and McCoy (5), seemed to indicate that cortical extract is without influence in counteracting the effects of bacterial toxins in normal animals. In the present work we studied the effect of cortical extracts on the course of bacterial intoxications in guinea pigs to which various doses of toxins were administered and the more chronic effects were observed.

[1] Aided in part by a grant from the Therapeutic Research Committee of the American Medical Association.

EXPERIMENTS

The cortico-adrenal extract used, in the experiments reported here, was prepared in our laboratory according to the method of Swingle and Pfiffner (11), (12). One cc. of the extract represented 30 grams of fresh bovine adrenal glands. Bacterial exotoxins (diphtheria, botulinus, and tetanus toxins) were chosen as the intoxicating agents and the M.L.D. was determined and checked routinely at different intervals of time. The guinea pigs were obtained, for the most part, from our own animal quarters from a mixed stock. The weights of the guinea pigs used in these experiments varied from 200 to 400 grams; but in each series of experiments, the guinea pigs were practically of the same age and weight, and in several series, litter mates were employed. Nevertheless, individual variation in response was considerable. In the experiments with diphtheria toxin, 171 guinea pigs were sacrificed; 72 animals were given botulinus toxin and 18 pigs were employed in the experiments on tetanus toxin.

Diphtheria Toxin

The diphtheria toxin (No. 312 M.L.D. 600 Mulford) was obtained from Dr. J. C. Munch of the Mulford Biological Laboratories, to whom we are

TABLE 1. EFFECT OF CORTICO-ADRENAL EXTRACT IN THE GUINEA PIG AFTER INJECTION OF 3 M.L.D. OF DIPHTHERIA TOXIN

Group	Pig No.	Weight grams	Type of Treatment	Result
I	1	265	Cortico-adrenal Extract 1.0 cc. intramuscularly at 12 hour intervals	Died 25 hours
	2	240		" 29 "
	3	260		" 33 "
	4	310		" 29 "
	5	285		" 37 "
	6	265		" 30 "
II	7	305	Normal Saline 1.0 cc. intramuscularly at 12 hour intervals	Died 38 hours
	8	255		" 32 "
	9	265		" 31 "
	10	290		" 28 "
	11	260		" 36 "
	12	300		" 31 "
III	13	270	Untreated Controls	Died 26 hours
	14	275		" 34 "
	15	315		" 29 "
	16	265		" 28 "
	17	280		" 21 "
	18	270		" 30 "

deeply indebted. Separate dilutions of the toxin were made in normal saline for each series of experiments and at varying time intervals the toxin was checked as to the M.L.D.

Experiment 1. A series of 18 guinea pigs of approximately the same age and weight were divided into 3 groups of 6 each. All the animals were given 1 cc. of a 1-200 dilution of diphtheria toxin subcutaneously, equivalent to 3 M.L.D. The results are given in table 1.

It may be seen that no protection was afforded by the extract administered to

guinea pigs given 3 M.L.D. of diphtheria toxin. At autopsy, the adrenal glands of both the experimental and control animals showed marked hyperemia and hemorrhage.

Experiment 2. A series of 18 guinea pigs were employed to study the effect of the extract on the course of the intoxication produced by 1 M.L.D. of diphtheria toxin. The same procedure was carried out as in the previous experiment. No appreciable benefit could be seen from the administration of the cortical extract.

Another series of 15 guinea pigs was divided into 3 groups as previously mentioned. One group of pigs in the experiment received 1 cc. of extract daily for 2 days prior to the inoculation with 1 M.L.D. of diphtheria toxin and daily thereafter until death. The results are shown in table 2.

TABLE 2. EFFECT OF CORTICO-ADRENAL EXTRACT ON GUINEA PIGS AFTER INJECTION OF 1 M.L.D. OF DIPHTHERIA TOXIN

Group	Pig No.	Type of Treatment	Results
I	37	Cortico-adrenal Extract	Died 61 hours
	38		" 70 "
	39		" 83 "
	40		Died in paralysis 23 days
	41		" " 25 "
II	42	Normal Saline	Died 44 hours
	43		" 97 "
	44		" 54 "
	45		Died in paralysis 21 days
	46		Died 65 hours
III	47	Untreated Controls	Died 93 hours
	48		" 60 "
	49		" 74 "
	50		" in paralysis 19 days
	51		" " " 20 "

No definite protection was evident in the experimental animals which died in the acute stage. However, it was noted that 2 of the extract-treated animals which survived the acute toxemia and later died of post-diphtheritic paralysis (toxin effect), lived about 4 to 5 days longer than did similar controls.

Experiment 3. To further study the effect of the extract on the toxon of diphtheria, 30 guinea pigs were divided into 5 groups of 6 each, and treated as indicated in table 3. Each animal received about 0.8 M.L.D. (1 cc. of a 1-750 dilution of diphtheria toxin).

It appeared from this experiment that the cortical extract exerted a beneficial action in prolonging the life of those guinea pigs which had survived the acute stage of diphtheria intoxication. Three further series of such experiments were performed, bringing the total of guinea pigs which died of post-diphtheritic paralysis up to 66. Of these, the cortico-adrenal extract-treated pigs (16 in number) lived an average of 4 to 5 days longer than did the untreated controls. Cortical extract diluted in water instead of saline, acted in the same manner as did the cortical extract in saline. The saline-treated pigs lived on an average of 2 days longer than the untreated controls. The guinea pigs receiving inactivated cortical extract succumbed about 4 days earlier than

TABLE 3. EFFECT OF CORTICO-ADRENAL EXTRACT ON SURVIVAL PERIOD OF GUINEA PIGS AFTER INJECTION OF 0.8 M.L.D. OF DIPHTHERIA TOXIN

Group	Pig No.	Type of Treatment	Toxic Phenomena	Lethal in Days
I	52	Cortico-adrenal Extract, in saline, 1 cc. daily	Post-diphtheritic paralysis	24
	53		Acute toxemia	5
	54		" "	4
	55		Post-diphtheritic paralysis	27
	56		" " "	32
	57		" " "	21
II	58	Cort. adrenal Ext., in water, 1 cc. daily	Acute toxemia	5
	59		Post-diphtheritic paralysis	20
	60		" " "	26
	61		" " "	25
	62		" " "	21
	63		" " "	31
III	64	Cort. adrenal Ext. inactivated by heat, 1 cc. daily	Post-diphtheritic paralysis	18
	65		" " "	14
	66		" " "	19
	67		" " "	20
	68		Acute toxemia	6
	69		" "	4
IV	70	Normal Saline	Post-diphtheritic paralysis	22
	71		" " "	23
	72		" " "	23
	73		Acute toxemia	5
	74		Post-diphtheritic paralysis	21
	75		Acute toxemia	4
V	76	Untreated Controls	Post-diphtheritic paralysis	16
	77		Acute toxemia	4
	78		Post-diphtheritic paralysis	24
	79		" " "	19
	80		" " "	23
	81		Acute toxemia	5

did the controls, indicating a possible toxic substance in the heated extract. No difference in survival time was observed in those pigs which died during the acute stage of diphtheritic toxemia.

Botulinus Toxin

The botulinus toxin was prepared in the Bacteriology Department of the School of Medicine from the 8 AB Botulinus type, Zinsser strain. It was standardized and found to have 10,000 M.L.D. per cc.

Experiments performed on 15 guinea pigs, after injection of 10 M.L.D., failed to reveal any marked differences in the survival time between the extract-treated animals and the controls. Experiments with 2 M.L.D. on another 15 guinea pigs, likewise did not disclose that any significant protection had been afforded by the extract.

Thirty male guinea pigs of approximately the same age and weight were divided into 3 groups, treated as shown in table 4.

Of the 12 extract-treated animals, 4 survived and the remaining 8 lived an average of 89 hours following the administration of the toxin; of the 12 saline controls, 1 pig survived and the remaining 11 lived an average of 78

TABLE 4. EFFECT OF CORTICO-ADRENAL EXTRACT ON GUINEA PIGS AFTER INJECTION OF 1 M.L.D. OF BOTULINUS TOXIN

Group	Pig No.	Type of Treatment	Results
I	31	Cortico-adrenal Extract, 1 cc. 4 hours before injection of toxin and daily thereafter	Died 72 hours
	32		Survived
	33		Died 91 hours
	34		" 93 "
	35		" 74 "
	36		Survived
	37		Died 118 hours
	38		" 96 "
	39		" 83 "
	40		" 90 "
	41		Survived
	42		"
II	43	Normal Saline	Died 50 hours
	44		" 59 "
	45		" 82 "
	46		" 62 "
	47		" 87 "
	48		" 123 "
	49		" 72 "
	50		" 80 "
	51		Survived
	52		Died 91 hours
	53		" 81 "
	54		" 76 "
III	55	Untreated Controls	Died 64 hours
	56		" 62 "
	57		" 86 "
	58		" 70 "
	59		" 102 "
	60		" 64 "

hours after toxin administration. The untreated controls lived 74.5 hours (average) following the injection of 1 M.L.D. of toxin.

A series of 12 guinea pigs were subjected to 0.7 M.L.D. of botulinus toxin, but all survived and no results could be obtained.

Tetanus Toxin

The tetanus toxin was prepared from the organism isolated from a fatal case of tetanus poisoning (Autopsy #1676) by Mr. N. Duffett of the Bacteriology Department. After the M.L.D. had been determined, 18 guinea pigs were divided into 3 groups of 6 each. After injection of 1 M.L.D. of toxin subcutaneously, Group I received 1 cc. of extract daily; Group II received saline, and Group III served as a control. The findings are given in table 5.

The extract-treated pigs appeared to do worse than did the controls. None of the experimental animals recovered, whereas one saline-treated and 2 untreated controls survived. The average length of life after injection of the toxin for the extract-treated animals was 4.3 days, as compared to 5.4 days for the controls.

DISCUSSION

The findings reported here regarding the effect of cortical extract on the response to lethal doses of diphtheria toxin in guinea pigs, are confirmatory

to those of Scott, Bradford, Hartman, and McCoy (5). The acute diphtheritic toxemia is probably too overwhelming for the extract to be of much value. However, in the guinea pigs which survive this acute stage, another process becomes manifest due to the toxon of diphtheria, i.e. post-diphtheritic paralysis and during this stage, the administration of cortical extract appears to be of benefit in prolonging the life of the animals. The mechanism for this action is not known. Does the cortical extract increase the resistance of the organism to the toxon or does the extract exert a stimulating action on the nervous system and thus delay the onset of the peripheral neuritis with its paralysis? This question cannot be answered at present. It has long been recognized that diphtheria toxin has somewhat of a special affinity for the adrenal gland. Belding and Wyman (13) reported that adrenalectomized rats had a greater

TABLE 5. EFFECT OF CORTICO-ADRENAL EXTRACT ON GUINEA PIGS AFTER INJECTION OF 1 M.L.D. OF TETANUS TOXIN

Group	Pig No.	Weight grams	Type of Treatment	Result
I	1	360	Cortico-adrenal Ext.	Died 4 days
	2	325		" 6 "
	3	348		" 4 "
	4	382		" 4 "
	5	365		" 3 "
	6	350		" 5 "
II	7	385	Normal Saline	Died 8 days
	8	320		Survived
	9	345		Died 5 days
	10	340		" 4 "
	11	365		" 4 "
	12	325		" 6 "
III	13	390	Untreated Controls	Died 9 days
	14	320		" 4 "
	15	345		Survived
	16	365		Died 5 days
	17	360		" 4 "
	18	330		Survived

susceptibility to diphtheria toxin and that the elimination of the toxin from the blood was much slower in the adrenalectomized rats than in controls. However, no reports have appeared to show that the injection of cortico-adrenal extracts increases the resistance of animals to diphtheria toxin.

The action of cortical extracts on the neuromuscular system appears to be more attractive as an explanation for the experimental findings in this paper. Nerve-muscle preparations from adrenalectomized rats were found by Gans and Miley (14) to become more easily fatigued than did normal nerve-muscle preparations. Hartman, Beck and Thorn (15) observed stimulating effects of cortical extracts on the nervous system in human subjects. These workers also reported an increase in the power of muscle to work and an increase in the resistance to fatigue (16). In the experiments reported in this paper, a beneficial effect was noted in post-diphtheritic paralysis and in the paralysis caused by botulinus toxin. In both of these conditions, the peripheral nervous system is involved; in diphtheria, the peripheral nerves, and in botulinus

poisoning, the motor end plates. It is quite possible that the action of the cortical extract in these conditions is related to its action on the neuromuscular system. In tetanus intoxication, the activity of the synapses in the anterior horn of the cord is apparently stimulated and here the cortical extract appeared to increase the severity of the symptoms. More work on this phase of the problem is contemplated before any definite conclusions can be reached.

SUMMARY

The effect of cortico-adrenal extract on the course of diphtheria, botulinus, and tetanus intoxication in guinea pigs was studied.

In a series of 105 guinea pigs which received lethal doses of diphtheria toxin and which died in acute toxemia, no protective action could be demonstrated from the administration of cortico-adrenal extract.

Of the 66 guinea pigs which survived the acute stage and succumbed to post-diphtheritic paralysis, the cortical extract prolonged the lives of the experimental animals on an average of 4 to 5 days over those of untreated controls.

In a series of 30 guinea pigs which received 2 or more M.L.D. of botulinus toxin, no definite benefit was shown from cortico-adrenal extract administration. In 30 guinea pigs which were inoculated with 1 M.L.D., a slight but definite protective effect was noted in those pigs which were injected with cortico-adrenal extract. The number of pigs which lived and the period of survival was greater in the extract-treated series than in the controls.

No therapeutic effect could be demonstrated from the use of cortico-adrenal extract in guinea pigs with tetanus intoxication. A slight synergistic action was apparent between the tetanus toxin and the cortical extract.

REFERENCES

1. Lewis, J. T.: Am. J. Physiol. **64:** 506. 1923.
2. Jaffe, H. L., and D. Marine: J. Infect. Dis. **35:** 334. 1924.
3. Scott, W. J. M.: J. Exper. Med. **39:** 457. 1924.
4. Perla, D., and J. M. Marmorston-Gottesman: J. Exper. Med. **47:** 723. 1928.
5. Scott, W. J. M., and W. L. Bradford: Proc. Soc. Exper. Biol. & Med. **28:** 428. 1931.
6. Perla, D., and J. M. Marmorston-Gottesman: Proc. Soc. Exper. Biol. & Med. **28:** 475. 1931.
7. Scott, W. J. M., W. L. Bradford, F. A. Hartman, and O. R. McCoy: Endocrinology, **17:** 529. 1933.
8. Wenner, W. F.: Arch. Otolaryng. **17:** 774. 1933.
9. Whitehead, R. W., and C. Smith: Proc. Soc. Exper. Biol. & Med. **29:** 672. 1932.
10. Wenner, W. F., and A. J. Cone: Arch. Otolaryng. **20:** 178. 1934.
11. Swingle, W. W., and J. J. Pfiffner: Am. J. Physiol. **96:** 164. 1931.
12. Pfiffner, J. J., and W. W. Swingle: Am. J. Physiol. **96:** 180. 1931.
13. Belding, D. L., and L. C. Wyman: Am. J. Physiol. **78:** 50. 1926.
14. Gans, H. M., and H. H. Miley: Am. J. Physiol. **82:** 1. 1927.
15. Hartman, F. A., G. M. Beck, and G. W. Thorn: J. Nerv. & Ment. Dis. **77:** 1. 1933.
16. Hartman, F. A., and G. W. Thorn: Proc. Soc. Exper. Biol. & Med. **29:** 48. 1931.

REID HUNT REACTION AND THE THYROTROPIC HORMONE[1]

HANS WIESBADER

From the Department of Obstetrics and Gynecology, College of Physicians and Surgeons, Columbia University

NEW YORK CITY

Reid Hunt's observation that white mice treated with thyroid substance showed an increase in their resistance against acetonitril, gave us a very suitable method for the study of thyroid disturbances. Hunt considers the protective action as due to acceleration of the oxidation of acetonitril, formic acid and thiocyanate. In all probability the mechanism of detoxication is more complicated. A diminution of the effect of the poison results. Numerous writers (Ghedini, Gellhorn, Miura, Reid Hunt, von Bergmann and others) found that this reaction was not specific for the thyroid but could also be produced by other organ extracts. With the increase in knowledge of the endocrines the reason for this peculiar behavior becomes clearer. It offers an explanation of why the administration of the serum of pregnant women or anterior pituitary gland extracts increases the resistance to acetonitril. Thyroxin and the thyrotropic hormone are responsible for the elevated Reid Hunt reaction. Animal tests have shown unquestionable histological changes in the thyroid after the administration of the serum of pregnant women (Schenk) in the form of stimulation. A further proof for the activation of the thyroid during pregnancy was given by Eufinger and Wiesbader who demonstrated the acceleration of metamorphosis of frog larvae after the addition of serum of pregnant women.

The following experiments show the effect of serum of normal subjects, serum of pregnant women in the ninth and tenth months of pregnancy, of anterior pituitary-like hormones (follutein) and the thyrotropic hormone.

EXPERIMENTS

Male white mice weighing 20 grams, which had been on a standard diet of oats and water for 4 weeks were used. On 3 successive days the animals were injected with 0.5 cc. of either normal serum or serum of pregnancy, 0.4 cc. of follutein or thyrotropic hormone, and 24 hours after the last dose were injected with increasing doses of acetonitril. Five mice were used for every test. The first animal was given the lethal dose which was always determined on untreated controls and which was 0.6 mg. of acetonitril per gram of body weight. The animals following were given successive increases of 0.1 mg. acetonitril per gram of weight. The result was noted 24 hours after the last injection.

Serum from normal subjects. Mice given serum from 10 normal subjects showed no increase in their resistance to acetonitril (lethal dose 0.6 mg. per gram of body weight). It seems that in the blood of normal human beings

[1] This work was supported by a grant from E. R. Squibb and Sons.

TABLE 1.

No.	Pregnancy	Lethal dose in mg. per gram body weight	% increase over 0.6 mg.
1		0.8	33
2		0.8	33
3		0.9	50
4		0.9	50
5	9th month	0.9	50
6		0.8	33
7		0.9	50
8		0.9	50
9		0.8	33
10		0.8	33
1		0.9	50
2		0.9	50
3		1.0	66
4		0.9	50
5	10th month	1.0	66
6		1.0	66
7		0.9	50
8		0.9	50
9		0.9	50
10		0.9	50

the amounts of thyroid and thyrotropic hormone are too small to give a direct Reid Hunt reaction. Oehme and Paal made use of the indirect Reid Hunt reaction in order to find the thyrotropic hormone in the blood of normal human beings. They injected into rabbits fractionated human serum which had previously had no effect in the direct Hunt reaction, and demonstrated 24 hours later an increase of the acetonitril resistance in the rabbit serum which was equal to the lethal dose.

Pregnancy serum. Serum was obtained from 20 normal women in their ninth and tenth lunar months of pregnancy. Injection of 0.5 cc. of this serum was made in male mice on three consecutive days. As shown in table 1 the resistance to acetonitril was increased over the usual lethal dose of 0.6 mg. per gram.

Pregnancy urine (follutein). This preparation was injected in doses of 0.4 cc. (equal to 6 mouse units). As shown in table 2, the acetonitril re-

TABLE 2. MINIMAL LETHAL DOSES OF ACETONITRIL IN MICE TREATED WITH FOLLUTEIN AND WITH THYROTROPIC HORMONE

Dose of acetonitril mg./gram body weight	Mice treated with 6 M.U. follutein			Mice treated with 7.2 M.U. thyrotropic hormone		
	Survived	Died	% increase over lethal dose of 0.6 mg.	Survived	Died	% increase over lethal dose of 0.6 mg.
0.6	5	0	0	5	0	0
0.7	5	0	16	5	0	16
0.8	4	1	33	5	0	33
0.9	2	3	50	3	2	50
1.0	0	5	66	2	3	66
1.1				0	5	83

sistance of the mice were increased 66 per cent by prolan A and B, a lethal dose of 1.0 mg as compared with the normal of 0.6 mg. being found.

Thyrotropic hormone. This preparation was injected in doses of 0.6 cc., equal to 7.2 mouse units. The data in table 2 show that the acetonitril resistance of the mice was increased by the thyrotropic hormone and that the lethal dose was 1.1 mg., an increase of 83 per cent over the normal lethal dose of 0.6 mg.

CONCLUSIONS

Normal blood serum does not increase the resistance of mice to acetonitril poisoning. The resistance of mice to acetonitril is increased by preliminary treatment of the animals with blood serum from pregnant women; with follutein, an anterior pituitary-like hormone extracted from pregnancy urine; and with thyrotropic hormone from the anterior pituitary.

I am indebted to Professor Benjamin P. Watson for constant encouragement and advice, and for his placing the large material of the Sloane Hospital for Women and the Vanderbilt Clinic at my disposal.

REFERENCES

Bruehl, R.: Klin. Wchnschr. **8:** 254. 1929.
Dallera, N.: Ann. di ostet. e. ginec. **53:** 831. 1931.
Engelhorn, E.: Schilddruese und weibliche Geschlectsorgane, Erlangen, 1912, Junger and Sohn.
Eufinger, H., H. Wiesbader, and L. Focsaneanu: Arch. f. Gynäk. **136:** 12. 1929.
Ghedini, G.: Wien. Klin. Wchnschr. **24:** 736. 1911.
Herold, L.: Arch. f. Gynäk. **154:** 256. 1933.
Hunt, Reid: J. Biol. Chem. **1:** 33. 1905-1906.
Hunt, Reid: Brit. M. J. **2:** 1788. 1906.
Hunt, Reid: Am. J. Physiol. **63:** 257. 1922-1923.
Oehme, C. and H. Paal: Ergebn. d. inn. Med. u. Kinderh. **44:** 214. 1932.
Paal, H.: Klin. Wchnschr. **10:** 2172. 1931.
Schenk, F.: Zentralbl. f. Gynäk. **57:** 2232. 1933.
von Bergmann, G.: Med. Klin. **24:** 844. 1928.
von Bergmann, G., and M. Goldner: Ztschr. f. klin. Med. **108:** 100. 1928.

AN INVESTIGATION OF THE HORMONE CONTENT OF SALIVA

ABNER I. WEISMAN AND CHARLES C. YERBURY

From the Gynecological Division of the Endocrine Clinic of the Metropolitan Hospital, and the Department of Obstetrics and Gynecology of the New York Homeopathic Medical College and Flower-Fifth Avenue Hospital

NEW YORK CITY

Since the first finding of the anterior pituitary-like (AP-L) and the œstrus-producing hormones in urine of pregnancy in 1926 the hormones have been found in various other body fluids and tissues. The œstrus-producing hormone has been identified in blood, amniotic fluid, placenta, corpus lutem, feces, milk, bile and liver. The AP-L hormone besides being found in urine has been extracted from placenta and identified in blood.[1] It is also known that the hormones are not specific for pregnancy since in both non-pregnant females and normal males both hormones can be demonstrated in urine and blood in smaller amounts (1), (2), (3).

The close relationship of the salivary secretion to the blood stream and the recognized constituents of saliva are generally known. It is generally conceded that pregnancy in rare cases is the underlying cause for excessive salivation and that hyperemesis gravidarum and excessive salivation of pregnancy are due to a hormone imbalance at that period when the hormones are found in urine in varying amounts (4), (5). Thus, the widespread presence of the sex hormones in the body fluids, the close association of saliva to the blood, the linking of salivation with pregnancy and the fact that no hormone study of saliva had yet been made caused us to examine various salivary secretions for the presence of the sex hormones.

METHODS

The experiments were performed in the following manner. Two daily subcutaneous injections of 0.4 cc. each of (a) male saliva, (b) saliva of an adolescent female, (c) saliva of a married non-pregnant female, (d) different salivas of each of four pregnant women in the third, fourth, fifth and sixth months of gestation, (e) saliva of a pregnant woman with excessive salivary secretion and no other toxic manifestation or finding, were given to immature female white mice seventeen days old. As controls, 0.4 cc. of human pregnancy urine was injected twice daily into a ninth group of mice. Five animals were used for each material tested. Five animals were also kept without injection, as a second group of controls. The mice were given the injections for three days and all the animals were killed on the morning of the fourth day. The entire experiment was repeated using saliva concentrated for AP-L hormone to 10 times the original amount, using a crude method of extraction (6).

[1] Complete reference to all the preceding reports may be found in Chapter III Vol. 1, of Endocrine Medicine by William Engelbach. 1932.

The ovaries were removed and examined without staining under the low power microscope lens. The ovaries of all the non-injected control animals were pale and clear, showing no evidence of corpora lutea. The group of animals injected with the known positive human pregnancy urine all showed the gross characteristic signs of the Aschheim-Zondek reaction, swelling, turgescence, increase in size of the ovaries, and blood-point hemorrhages. The group of mice injected with all the various types of salivas both whole saliva and the extracted saliva showed no positive reaction either grossly or microscopically, and were no different from the non-injected group.

Two daily subcutaneous injections of 0.5 cc. each of salivas from the same sources were given to mature female white rats previously castrated and thoroughly tested by us for the œstrus reaction. A similar control of human pregnancy urine was injected twice daily to a ninth group of rats. Five animals were also used for each material tested and a group of animals were also kept without injection as a second group of controls. The rats were given injections for three days and vaginal smears were taken at 24, 48, 72, and 96 hour periods after the first injection. The entire experiment was repeated using saliva concentrated for the œstrus-producing hormones to ten times the original strength of the saliva (7). Smears were stained with eosin-haematoxylin and carefully examined under the microscope. The vaginal smears of the group of animals injected with the known positive human pregnancy urine were for the most part markedly positive at the 96 hour readings, with nothing but desquamated cornified vaginal epithelium seen in the smears. The non-injected control animals as well as the animals injected with the whole salivas and the concentrated salivas gave smears which were typically negative.

DISCUSSION

In pregnancy, the urine and blood have a much greater hormone content than otherwise and with the urine acting as the excretory medium of the hormone above the threshold value of the blood, it hardly seems necessary that the saliva should remove any appreciable amount of hormone from the blood stream when the hormone content is probably maintained at a definite level of concentration by the excretion of urine.

SUMMARY

Salivary secretions tested by the Aschheim-Zondek test and the vaginal smear test contained no anterior pituitary-like hormone or œstrus-producing hormones, or too small an amount to give positive reactions.

REFERENCES

1. Frank, R. T., M.A. Goldberger, and F. Spielman: Proc. Soc. Exper. Biol. & Med. **28:** 999. 1931.
2. Laqueur, E., E. Dingemanse, P. C. Hart, and S. E. de Jongh: VI. Mitteiling. Klin. Wchnschr. **6:** 1859. 1927.
3. Frank, R. T., and M. A. Goldberger: Proc. Soc. Exper. Biol. & Med. **25:** 476. 1928.
4. Smith, G., and O. Smith: Am. J. Physiol. **107:** 128. 1934.
5. Hirst, J. C.: J.A.M.A. **66:** 645. 1916.
6. Katzman, P. A., and E. A. Doisy: Proc. Soc. Exper. Biol. & Med. **31:** 188. 1933.
7. Harrow, B., and C. P. Sherwin: The Chemistry of Hormones. p. 157, Williams and Wilkins Co., Baltimore, 1934.

CLINICAL AND LABORATORY NOTES

OBESITY AND OCULAR SYMPTOMS IN MENTALLY ALERT CHILDREN DUE TO HYPOTHYROIDISM

JOS. P. COSTELLO

In all hypothyroid patients metabolism is decreased resulting in a storing up within the tissues of the body an excess of energy producing substances. It seems that in such pathological conditions less food is required to complete metabolic processes than in the normal individual. As a result all patients with definite thyroid deficiency will develop obesity.

It is generally believed that when there is a definite thyroid deficiency the mentality will be affected. Either the mind of a child will not develop normally such as is seen in cretinism, or if symptoms of hypothyroidism develop later in life mentality will become less and less acute as the disease progresses. Mentality is not always thus affected in hypothyroid states. In fact many children with definite symptoms are of normal mentality, and some are unusually bright. It is the bright alert child, with a tendency toward obesity that generally lacks thyroid.

In all thyroid conditions the eyes of the patient show some abnormal condition. In the hyperthyroid we have the exopthalmus and in the hypothyroid child there is usually found myopia with or without astigmatism.

A delay in ossification and growth of bone does occur in certain types of thyroid deficiency but not in the type being discussed here.

Dryness of the skin, dry thin hair, and the so-called pseudoedema is also lacking in the type of a patient described here.

The syndrome seen frequently by every pediatrician is as follows. A bright, alert little girl gives a history of having been fat all of her life. As a baby development was normal. She sat up, teethed and walked at the normal age. Mentally she has always been bright and has taken a keen interest in all of her surroundings. At the attendance of school it was noticed that she complained of her eyes tiring and pain in the eyes after school work. The appetite is good but to use the mother's expression "everything she eats turns to fat." Her endurance is embarrassed by her obesity. Such patients continue to gain in spite of the fact that low caloric diets have been given.

An examination shows a well developed little girl considerably overweight for her height, although the height for the age is normal. The fat is evenly distributed over the body and there is no padding such as is seen in the pituitary types. An examination of the eyes will show some degree of myopia. Frequently the tonsils and adenoids are enlarged but a careful history will show that they seldom have been infected. Tooth development is normal. The thyroid is not enlarged. The thymus does not show an enlargement in both anterior and posterior positions when x-rayed. The heart is slower than normal but not otherwise affected. The abdomen is large. Liver and spleen are not palpable. The genitalia show normal develop-

ment. The skin is normal in texture and there is no abnormality of hair growth. There is no adenopathy. Reflexes are present and normal.

When the basal metabolic rate is determined on such a patient, one is surprised to find a minus 14 to 26. The low basal is entirely out of proportion to the apparent health and mentality of the individual. Likewise, there is a drop in creatinine and not infrequently a drop in the blood sugar.

Therapy. It was thought, at the onset of my clinical observation of such cases, that the entire picture might be one of avitamintosis. However, when vitamin B was added to the diet in large quantities, no appreciable change in the patient was noticed.

Thyroid therapy in this condition is most valuable. Small doses should be given at the onset as such patients seem to have a low tolerance for thyroxin and will develop toxic symptoms if large doses are given during the early stages of treatment. The first noticeable signs of improvement will be found in the eyes. The myopia will be greatly improved but seldom do the eyes return to normality. The loss in weight is a slow process because of the small dosage. Months of treatment are required before such children show an appreciable loss. The low blood sugar and creatinine when present in such patients responds at once, but will remain lower than normal in spite of treatment.

Conclusions. (1) Hypothyroidism in a growing child does not always affect mentality. (2) Obesity is a constant sign of hypothyroidism; growth is not always affected. (3) Myopia or some eye disturbance is always found in the above clinical picture. (4) Thyroid by mouth greatly improves the syndrome but does not completely cure the patient.

Lister Building.
St. Louis, Missouri.

VITAMIN E AND THE GONADS

WILLIAM SAPHIR*

Since Evans' discovery of the antisterility factor or Vitamin E, which is abundantly present in the wheat germ, it has been shown conclusively that this factor is essential for normal gestation in rats. Lack of Vitamin E in the diet of pregnant rats invariably leads to placental separation and death of the foetus. A similar rôle of this factor in the normal gestation of another species of animals is indicated by the observations of Ewing[1] on the high incidence of abortion in hogs deprived of Vitamin E. A still wider application of the same principle seems apparent from the report[2] that abundant Vitamin E has been found in the "royal jelly," the food juice of the future queen-bee, while the food of the sterile working bee is lacking in this respect.

As regards the indispensability of Vitamin E in human nutrition, Balfour and Talpade[3] have reported that in Southern India, where milled rice is the staple diet, the incidence of premature births and stillbirths is three times greater than in Northern India, where whole wheat is widely used. Although it is difficult to conceive of a similar deprivation of Vitamin E occurring in humans living on the mixed diets of western peoples, Vogt Mueller,[4] Watson,[5] and McGonigle[6] were successful in treating a large percentage of their cases of habitual abortion with wheat germ oil. It seems more reasonable to ascribe these beneficial results to an excess of Vitamin E rather than to the correction of an absolute deficiency.

A number of commercial preparations of wheat germ oil, biologically assayed for Vitamin E content, have recently appeared upon the market. There seems to be an increasing clinical tendency to employ these preparations not only in the treatment of recurrent abortion, but also in cases of primary sterility in the female. In contrast with the observations on habitual abortion, the clinical reports of the therapeutic results in primary sterility are not impressive. Furthermore, the gonadotropic action of wheat germ oil, which such results would imply, is disputed. Szarka[7] claimed to have observed an estrogenic activity of an ether extract of wheat germs in infantile rats. Olcott and Mattill,[8] however, failed to confirm these results; nor could they discern any effects upon the ovary.

The availability and commercial exploitation of new therapeutic products often leads to a period of overenthusiastic use of such materials, with a consequent neglect of older, time-tried methods. Because of this and of the foregoing discussion, it was thought worthwhile, at this time, to attempt to demonstrate a possible gonadotropic or estrogenic activity of amounts of Vitamin E, far in excess of ordinary nutritional requirements.

Parallel tests were made throughout on two commercial preparations,[9] as regards estrogenic, gonadotropic and luteinizing activity. The results were entirely negative and are summarized very briefly as follows:

* The Department of Metabolism and Endocrinology, Michael Reese Hospital, Chicago.

[1] Evans, H. M., Science **56**: 650, 1922. Ewing, P. V., The Hog Breeder **10**: 3, 1935.
[2] Hill, L., and Burdet, E. F., Nature (London) **130**: 340, 1932.
[3] Balfour, M. I., and Talpade, S. K., Indian Med. Gazette **67**: 601, 1932.
[4] Vogt Moeller, P. Acta obst. et gyn. Scandinav. **13**: 219, 1933.
[5] Watson, E. quoted in Editorial Ill. Med. J. **68**: 97, 1935.
[6] McGonigle, C. C. M., Ill. Med. J. **67**: 212, 1935.
[7] Szarka, A. Pflueger' Archiv. f.d.ges. Physiol. **223**: 657, 1930.
[8] Olcott, H. S., and Mattill, H. A., J. Biol. Chem. **104**: 423, 1934.
[9] Triticol (Pharmaceutical Specialties Co.) and Wheat Germ Oil (Double D Laboratories), kindly supplied by the respective firms. Both these products are said to be produced by cold pressing.

Estrogenic Action. Castrated adult female rats were injected intraperitoneally with successive amounts of wheat germ oil, until a total of 10 cc. had been administered. Repeated vaginal smears during and after treatment never showed vaginal cornification according to the criteria of Allen and Doisy.

Gonadotropic Action. Infantile female rats were injected intraperitoneally with successive amounts of wheat germ oil until a total of 8 cc. had been administered. In no instance did the ovaries show any increased activity as compared to control rats.

Luteinizing Action. Infantile female rats were first injected with a follicle stimulating urine extract (castrate human) after which wheat germ oil was administered. All the rats showed large Graafian follicles, but no luteinization.

Since animals deprived of Vitamin E may conceive and proceed through an initial period of normal gestation, it is not necessary to predicate a disturbance in the normal cycle of events which precede and follow conception. Nevertheless, the efficacy of Vitamin E in the treatment of habitual abortion might be supposed to depend upon a failure of the corpora lutea to persist for their usual period in Vitamin E deficiency. However, we have been unable to demonstrate any luteinizing action of this factor.

The more doubtful therapeutic effect of Vitamin E in primary sterility might be supposed, with greater reason, to depend on an estrogenic or gonadotropic action. Here again, no such activity could be demonstrated.

Summary. Commercial preparations of wheat germ oil, biologically assayed for Vitamin E content, failed to demonstrate any estrogenic, gonadotropic or luteinizing effects when tested on immature female, and normal and castrated adult female rats. From this limited series of experiments we have been unable to show any experimental basis for explaining the efficacy of Vitamin E in the treatment of habitual abortion. Our work yields no support for the claim that Vitamin E is of value in the treatment of primary sterility in the female.

I wish to thank Dr. Samuel Soskin for aid and direction.

SHORT STATURE OF PATIENT—A CAUSE OF ERROR IN PERFORMING PLUMMER'S TEST FOR QUADRICEPS WEAKNESS IN EXOPHTHALMIC GOITRE*

ISRAEL H. MARCUS

Muscular weakness is of frequent occurrence in exophthalmic goitre. Plummer[1] called attention to the diagnostic value and to the ease of demonstrating this myasthenia in the case of the quadriceps muscles. He suggested that the physician observe with what facility or difficulty the patient was able to mount upon the examining table, and that thus the physician could learn the strength of the quadriceps muscles. Jackson[2] considers the test as one of the four most important diagnostic signs in exophthalmic goitre. Many modifications of the test have been employed. The usual method of performing the test at the Jewish Hospital is the following: An ordinary chair is placed before the patient, the physician steadying the back of the chair with his hands. The patient is instructed to step up on to the chair, but while doing so not to touch the chair or any part of her body with her hands. The result is rated simply either negative or positive, depending on her ability or inability to thus get up on the chair. In most cases of exophthalmic goitre, of any degree of severity beyond the mildest, the patient will be unable to step up on the chair without the aid of her hands.

In watching many excellent physicians test patients for quadriceps weakness I found that the stature of the patient was completely disregarded, and in a search of the literature I found no warning mentioned anywhere concerning this matter. Yet it is obvious, if we but once think about it, that a patient of very small height will, because of the shortness of her legs, have more difficulty in mounting upon the chair or table than will a normal or long legged individual. Many a short patient has, as a result, been thought to have weak quadriceps and consequently hyperthyroidism, when no such myasthenia really existed and her fault lay only in her diminutive stature. It is therefore recommended that in testing such patients, if we would avoid this error, which is so frequently made, a proportionately low chair be employed.

Summary. Attention is called to the fact that patients of short stature, tested by Plummer's technic for quadriceps weakness, are apt to be mistakenly judged as having hyperthyroidism, unless a suitably low chair or table be employed in the test.

---

* From the Meyer A. Rabinowitz Medical Service of the Jewish Hospital of Brooklyn, Brooklyn, New York.

[1] Plummer, W. A.: St. Paul M.J. 18: 297, 1916.

[2] Jackson, A. S.: Goiter And Other Diseases Of The Thyroid Gland, Paul B. Hoeber, Inc., New York, 1926, page 101.

CURRENT ENDOCRINE LITERATURE

COLLABORATORS

Fuller Albright
I. Andrussier
Wayne J. Atwell
Israel Bram
John C. Burch
John C. Donaldson
H. B. Friedgood
Maurice H. Friedman
Murray B. Gordon
Arthur Grollman
Frank A. Hartman
E. C. Hamblen
Edward Larson
J. T. Lewis
Joseph M. Looney
Warren O. Nelson
J. P. Pratt
A. T. Rasmussen
Elmer L. Sevringhaus
L. B. Shpiner
D. L. Thomson
R. L. Zwemer

Atrophy of the adrenals and Addison's disease. Susman, W. To appear in this Journal.

A study is presented of 18 cases of adrenal atrophy observed personally together with a review of the literature. Atrophy of the adrenals as a cause of Addison's disease has a high incidence, especially in England. The incidence of such cases in the available necropsy series varied from 0.025 to 0.29%. The female incidence is appreciably higher than that of the male and the highest frequency is between 35 and 45 years of age. An accumulation, in the adrenal, of minor atrophic lesions may be causal in some cases, but a female sex factor appears to be prominent, etiologically.

The effect of gonadectomy on the thyroid gland in the guinea pig. Kippen, A. A., and L. Loeb. To appear in this Journal.

Extirpation of the gonads in sexually mature young guinea pigs leads to an increase in the proliferative activity of the thyroid gland during the first week following the operation. There are in addition other changes indicating stimulation of the thyroid gland; of especial interest among the latter is an increase in the number of phagocytes which invade the colloid of acini, without undergoing early degeneration, and which may persist therefore for a longer period of time. In young immature guinea pigs these changes are less definite than in older ones. The stimulation of the thyroid gland accomplished by these means is on the whole very moderate and comparable to the effects obtained by administration of iodine preparations to guinea pigs rather than to the stimulation effected by optimal amounts of anterior pituitary extracts. It is pointed out that the removal of the follicular hormone from a female guinea pig sets into motion not only a mechanism leading to an increase in the follicular growth and maturation-inducing hormone which represents a specific effect, but in addition it induces an increased production of the thyroid-stimulating hormone in the anterior pituitary, which must be considered as distinct from the hormone responsible for follicular growth and maturation; this second process represents, therefore, a nonspecific effect of gonadectomy on the anterior pituitary gland.

Chemical changes in the muscle of hypophysectomised toads. Marenzi, A. D. To appear in this Journal.

The condition of asthenia developed in totally hypophysectomized toads or in toads in which only the glandular lobe of the hypophysis has been removed is accompanied by modifications in the chemical constitution of skeletal muscle as shown by its progressive decreased content of glycogen, inorganic phosphorus, phosphocreatine and glutathione and by the reduced lactic acid production during tetanic excitation. These facts demonstrate that a muscular factor is involved in the asthenia, fatigability and diminished capacity to work shown by the muscle of hypophysectomized toads. Grafts or extract-injections of either glandular or intermedio-neural lobe of the hypophysis relieve condition of asthenia and

always produce an increase in the amount of all the above mentioned substances lowered by the ablation of the total hypophysis or of its glandular lobe alone.

Cachexia hypophyseopriva (Simmonds' disease) with thyroid and suprarenal insufficiency. Rose, E., and G. Weinstein. To appear in this Journal.

The principal features of cachexia hypophyseopriva are briefly reviewed. An additional case is described: the patient was a Syrian female of 44 whose symptoms began following a difficult delivery at the age of 33. She was first seen at the age of 41 at which time a diagnosis of myxedema was made. She was next seen 3½ years later in terminal coma. At this time there was high fever, azotemia, hypoglycemia and hemoconcentration. At necropsy marked atrophy, fibrosis and destruction of normal architecture were noted in the anterior pituitary, suprarenal cortices and thyroid (in the latter there was also dense round cell infiltration). The posterior pituitary and pancreas were not affected. Certain features of the case suggest that suprarenal failure may have been the immediate cause of death.

Pituitary cachexia (Simmonds' disease). Report of a case in which the patient failed to respond to various forms of endocrine therapy. Moehlig, R. C. To appear in this Journal.

A case of a fifteen year old girl is reported who showed all the signs and symptoms of pituitary cachexia, or Simmonds' disease. During this time she received the anterior pituitary-like principle of pregnancy urine (antuitrin S), suprarenal cortex extract (eschatin), insulin alone and with glucose, pituitary tablets, cod liver oil, sunshine, a high caloric diet, and forced feeding. Despite these various forms of endocrine therapy and supportive measures, the patient died 17 months after the initial symptoms began.

Arrhenoblastoma, case report. Baldwin, L. G., and J. A. Gafford. To appear in this Journal.

A case of arrhenoblastoma in a 24 year old negress showing typical masculine changes is reported. After removal of the ovarian tumor there was a gradual return of feminism. Another laparotomy was made 4½ months later for a uterine fibroid. The literature on the condition is reviewed and the tabulation of Buettner has been brought up to date by the inclusion of later cases.

The reaction of fish to sex hormones. Owen, S E. To appear in this Journal.

Fish of the bitterling species (Rhodeus amarus) were employed in the assay of various concentrations of the prolans, theelin, theelol and antuitrin-S. The results indicate that the female bitterling is refractive to solutions containing these endocrine substances up to fifty mouse units per liter of aquaria water. The male bitterling, while reactive to solutions of these endocrines at concentrations of half a mouse unit per liter of aquaria water and more, gives inconstant results. It is not believed that fish would accurately supplant mice in a quantitative Aschheim-Zondek assay as is employed for teratoma patients. Approximately 75% of fish show positive assay findings when urines from pregnant women are employed. To avoid the possibility of antihormone interference, animals should not be used repeatedly in assaying hormones.

Prostatic atrophy following testicular feeding. Myers, H. B., B. Vidgoff, G. H. Schade and W. C. Hunter. To appear in this Journal.

Fourteen healthy, adult male rats were fed desiccated beef testis in amount of 0.2 gm. each per day over a period of 100 or more days. This resulted in atrophic changes in the testes, prostate and seminal vesicle, particularly in the glandular elements.

The presence of an excess of "male" (comb-growth and prostate stimulating) hormone in virilism and pseudo-hermaphroditism. Levy Simpson, S., P. de Fremery and A. Macbeth. To appear in this Journal.

The urine of 11 women with virilism and 3 pseudo-hermaphrodites was investigated for the content of "male" (comb-growth and prostate stimulating) hormone; and also oestrogenic hormone. The amount of "male" hormone was measured by its effect on comb-growth in capons, and increase in the weight of the prostate of castrated rats; the 2 methods, when compared, gave similar results. An excess of male hormone was found in 4 of 7 women with an adreno-genital syndrome, 3 women with a Cushing's syndrome, and 2 or 3 girls with pseudo-hermaphroditism. No excess was found in the remaining

3 women with an adreno-genital syndrome, 1 woman over 50 years of age with an Achard-Thiers syndrome, and 1 pseudo-hermaphrodite aged 4. A marked excess of male hormone was found in 1 woman with an adrenogenital syndrome (adrenal hyperplasia), 1 woman with a Cushing's syndrome (adrenal carcinoma), and 1 pseudo-hermaphrodite (adrenal hyperplasia).

Migraine and ovarian deficiency. Glass, S. J., H. R. Catchpole and Betsy McKennon. To appear in this Journal.

Therapy of migraine has been universally disappointing, chiefly because definite causes of a mechanism for the migrainous seizure have never been demonstrated. A group of 10 young migrainous women with ovarian deficiency was studied from an endocrine standpoint. Quantitative hormone assays of the ovarian and pituitary sex hormones revealed a reversal of the normal ratio of estrin and prolan in the direction of increased prolan and decreased estrin output. Adequate estrogenic therapy tends to correct this hormonal imbalance by actual suppression of the prolan A secretion with concomitant relief of the migrainous symptoms in 80% of the cases. Prolan administration gave no relief or intensified the symptoms. The exact relationship of the hyperprolanism to the migraine is unknown; however, any migraine associated with ovarian deficiency lends itself to a rational control with estrogenic therapy.

The induction of an acid vaginal secretion in the immature macaque by injections of oestrin. Hall, B. V., and R. M. Lewis. To appear in this Journal.

In 6 immature macaques *p*H determinations proved the vaginal fluid to be definitely alkaline. Three monkeys were then injected over a period of 2 weeks with sufficient oestrin to cause reddening of the sexual skin and to produce hyperplasia of the vaginal mucosa. The vaginal fluid of the injected animals was found to be highly acid. An increase in the H-ion concentration of at least 2 *p*H units was observed in each case. After withdrawal of the hormone and the subsequent return of the vaginal mucosa to the condition normal for the immature animal, the vaginal fluid was found to return to its former alkalinity. In one animal the vaginal fluid showed *p*H 7.8 forty-eight hours before reddening of the sexual skin occurred. With the discoloration an extraordinarily rapid shift took place giving a *p*H of 5. The results of similar observations on children, briefly detailed, indicated that this response to the injection of oestrin may explain the favorable results which have been reported in the treatment of infantile gonorrheal vaginitis.

The effect of injections of antuitrin S on the sexually inactive male ground squirrel. Baker, B. L., and G. E. Johnson. To appear in this Journal.

The daily subcutaneous injection of 5 R. U. of antuitrin S for 12 or 16 days into sexually inactive, adult, male ground squirrels had the following effects: the testes enlarged, increased in weight and, in general, partially descended into the scrotum; the seminiferous tubules enlarged and lumina formed in them; spermatogenesis was induced in all animals injected with antuitrin S, spermatozoa were present in 4 individuals after 16 injections; the seminal vesicles, the prostate and the Cowper's glands increased in size and weight and underwent secretory differentiation.

Excretion of prolan is essential hypertension. Scarf, M., and S. L. Israel. To appear in this Journal.

The prolan excretion, basal metabolic rate, specific dynamic action of protein and the sugar tolerance curves of 12 patients with essential hypertension were studied in an attempt to evaluate the theory of the pituitary origin of the disorder. No significant data resulted from this investigation with the possible exception of the presence of prolan (follicle-stimulating, gonadotropic principle) in the urine of 2 patients. These 2 had the highest blood pressure levels of the series, their systolic readings being over 200 mm. of mercury. One of the two patients, also, demonstrated other evidences of pituitary hyperfunction, having a high basal metabolic rate and a lowered sugar tolerance.

The biologic effects of thymectomy. Accruing retardation in growth in succeeding generations. Einhorn, N. H., and L. G. Rowntree. To appear in this Journal.

Thymectomy in successive generations of parent rats repeated generation by generation retards the rate of growth in successive generations of the offspring during the early weeks of life. This retardation is more pronounced in the third than in the second generation.

On vaso-dilator fibres in the sympathetic, and on the effect of circulating adrenaline in augmenting the vascular response to sympathetic stimulation. Burn, J. H., J. Physiol. **75:** 1454. 1932.

The experiments were designed to investigate the presence of vaso-dilator fibres in the sympathetic system. The experiments were performed on dogs (number not stated) who were eviscerated, exsanguinated and then divided into two parts at the lower end of the thorax. The pulmonary circulation was maintained by perfusing the lungs with defibrinated blood from another dog, this fluid being driven by one chamber of a Dale-Schuster pump—and by ventilating them through the trachea with a mixture of oxygen and 5% carbon dioxide. The blood was then perfused through the abdominal aorta by the second chamber of the pump, thus ensuring circulation to the hind part. Records were obtained of the pressure in the abdominal aorta after stimulation of the lumbar sympathetic chain and also after the addition of adrenaline to the circulating blood. It was found (1) that in the perfused preparation, stimulation of the lumbar sympathetic chain in the dog leads in most cases to vaso-dilatation, (2) that the magnitude of the vascular response to sympathetic stimulation, whether constrictor or dilator, is increased by the presence of circulating adrenaline.—H. Freeman.

A comparison of the effects of sympathin and adrenine on the iris. Cannon, W. B., and A. Rosenblueth, Am. J. Physiol. **113:** 251. 1935.

The effects were recorded of the action of adrenine, hepatic sympathin and cardio-pulmonary sympathin on the nictitating membrane (n.m.) and on the iris of 14 cats under dial anesthesia. When the 3 stimulating agents evoked nearly equal responses of the n.m. in 7 cats with normal iris, adrenine caused a marked dilation of the pupil, cardiac sympathin a slight widening and hepatic sympathin only minor and inconsistent effects. In 6 of 7 cats severance of the circular muscle of the iris in 2 places increased the enlargement of the pupil elicited by sympathin. Sympathin from the cardiac source however was still more effective than that from the hepatic source. The tentative conclusions were that sympathin I was present in hepatic and absent in cardiac sympathin and that sympathin E unlike adrenine, effected not only the dilator (sympathetic innervation) but also the constrictor muscle (parasympathetic innervation) of the iris.—A. J. Derbyshire.

Fetal carbohydrate metabolism following adrenalectomy, insulin and glucose experiments on the mother. Corey, E. L., Am. J. Physiol. **113:** 450. 1935.

The concentrations of glycogen in the maternal and fetal livers and in the placenta and fetal muscles were determined after adrenalectomy, and injection of insulin and glucose in normal pregnant rats. Adrenalectomy (19 animals) produced a depletion of both maternal and fetal liver glycogen. Injection of cortico-adrenal extract (15 animals) produced no significant change. Injection of insulin (10 animals) caused decreases of glycogen similar to those observed following adrenalectomy. Injections of glucose (9 animals) resulted in increases in liver glycogen in both mother and fetus. No significant changes were found in the glycogen of the placenta or fetal muscle.—F. A. Hitchcock.

Subtotal bilateral adrenalectomy for hyperadrenalism (essential hypertension). DeCourcy, J. L., Ann. Surg. **100:** 310. 1934.

Assuming that the cause of essential hypertension is a hyperplasia of the medullary tissue of the adrenal glands occurring under constant sympathetic stimulation and resulting in the secretion of excessive amounts of adrenaline into the blood stream, the author concluded that the most logical treatment of the condition would be extirpation of the excessive amount of overactive glandular tissue. He performs the operation under spinal anesthesia and in two stages separated by an interval of about 2 weeks. The portion extirpated includes both medulla and cortex removed at a distance from the entrance of the blood vessels. The kidney is exposed by the usual incision and held down with a special retractor. After the adrenal has been stripped of all fat and overlying structures, the portion to be removed is clamped and excised and the denuded surface is covered with a continuous chromic suture. Throughout the operation the blood pressure is watched closely. If the fall is more than anticipated, ephedrine is given promptly, and if collapse occurs, saline solution with adrenaline is administered intravenously. This operation is considered by the author to be entirely safe. He has performed it in 8 cases (16 operations). In every case the blood pressure remained low after the operation. The average drop was from 70 to 90 points in the systolic pressure and from 40 to 50 points

in the diastolic pressure. As it is probable that the chromaffin system other than the adrenals acts as a storehouse for adrenaline, a slight rise in the blood pressure may occur after the patient returns to his normal activities and persist for several months. In severe cases no untoward symptoms have developed even when as much as three-fourths of each gland was removed. It is concluded that the hypertension may well be regarded as the result of an endocrine dyscrasia for which the adrenals are responsible.—Author's Abst.

Effects of adrenalin injection in moderate work. Dill, D. B., H. T. Edwards and R. H. deMeio, Am. J. Physiol. **111**: 9. 1935.

About 5,000 Cals. of energy were expended by a fasting man in 24 hours. The energy derived from protein was about 1.5% of the total. In the early stages of work about one-half the energy was derived from carbohydrate; in the last 2 hours, less than one-tenth. In this subject the injection of adrenalin in 3 portions of 1 cc. each had no effect on protein metabolism, but increased the carbohydrate utilization more than ½ over the corresponding period in the control experiments. Small quantities of adrenalin produced smaller results. In other subjects the injection of adrenalin had an effect smaller in magnitude but of the same sign. Further evidence that adrenalin, instead of impairing, facilitates the oxidation of carbohydrate in moderate activity is found in the fact that the excretion of acetone bodies was always less after adrenalin than in corresponding periods of control experiments. The effects of adrenalin on blood sugar and lactic acid in work are like those in rest. When the concentration of blood sugar has reached its maximum, lactic acid has begun to decline while the R.Q. has already returned to the level of control experiments.—Authors' Summary.

The exhaustability of the sympathin stores. Dye, J. A., Am. J. Physiol. **113**: 265. 1935.

In more than 5 cats under dial anesthesia the sensitized nictitating membrane (n.m.) and blood pressure (b.p.) were used as respectively remote and local indicators for the exhaustion of sympathin produced by stimulating either splanchnic nerve. Both adrenals were tied off. Repeated brief stimulations failed to exhaust the stores of sympathin. Continuous faradic stimulation of a rate of 15/sec resulted in a loss of response of the n.m. in 1-2 hrs. while the elevation of b.p. was still present after 2 hrs. Direct stimulation of the cervical sympathetic did not exhaust the n.m. for 3 hrs. Exhaustion of sympathin to the threshold level for remote stimulations (n.m.) occurs earlier than that for its local effects (b.p.). Presumably sympathin stores are not exhausted by normal bodily functions.—A. J. Derbyshire.

Studies on the adrenal. X. Experimental studies on replacement therapy in adrenal cortical insufficiency. Grollman, A., and W. M. Firor, Bull. Johns Hopkins Hosp. **57**: 281. 1935.

The adrenal cortical hormone is more effective therapeutically when administered orally than when given by parenteral injection. Two preparations are described which served as complete replacement therapy when administered orally to dogs and rats, bilaterally adrenalectomized at one sitting. These preparations consisted of a charcoal adsorbate, previously described by the authors, and the product eluted from this charcoal combination. Available commercial preparations of the adrenal cortex were found to be devoid of any hormonal activity as manifested in their inability to prolong life in adrenalectomized animals.—A. G.

The mechanism of suprarenal insufficiency in the dog. Harrop, G. A., R. Ellsworth, L. J. Soffer, J. H. Trescher and A. Weinstein. Tr. A. Am. Physicians **48**: 221. 1933.

Experimenting on male adult dogs suprarenalectomized in two stages, there occurred on the withholding of injections of cortical extract, hemoconcentration, fall in blood pressure and in body temperature, and collapse of the circulation. This shock, in the dog at least, is construed to be the result of cortical deficit rather than disturbed function of the medulla. The authors discuss the question of the fate of the electrolytes and water lost from the blood plasma in adrenalectomized dogs. They resort to the possibility of increased excretion through the kidneys. They point out that progressive dehydration with reduction in volume of plasma and interstitial fluids interferes not only with circulation but with tissue respiration of metabolism and the proper transport of nutrients and waste material. The phenomena observed in the suprarenalectomized dogs and the complete repair occurring when cortical extract is given may be ascribed to the removal of an essential part of the normal electrolyte regulation. This would seem to render superfluous the theory long postulated that some toxic product of metabolism produces the symptoms following deprivation of suprarenal cortex.—I. B.

The relation of the suprarenal cortical hormone to nitrogen metabolism in experimental hyperthyroidism. Koelsche, G. A., and E. C. Kendall, Am. J. Physiol. **113:** 335. 1935.

The effect of cortical hormone on the loss of nitrogen resulting from the intravenous injection of 10 mg. of thyroxin on two consecutive days has been studied on six dogs. Experiments are reported on normal, unilaterally and bilaterally suprarenalectomized animals. In all cases the cortical hormone showed a sparing action against the increased loss of nitrogen resulting from the administration of thyroxin. It also decreased the systemic effects of thyroxin (increased pulse rate and blood urea). Bilaterally suprarenalectomized animals that received thyroxin showed an increase in the maintenance requirement of the cortical hormone. All dogs that survived bilateral suprarenalectomy for considerable time showed enlargement of the thymus. It is suggested that the magnitude of the increase in nitrogen excretion resulting from the administration of thyroxin may depend on the amount of available cortical hormone in the animal's body.—F. A. Hitchcock.

Effect of adrenalectomy on salt metabolism in rats. Rubin, M. I. and Elizabeth T. Krick, Proc. Soc. Exper. Biol. & Med. **31:** 228. 1933.

Adrenalectomized rats were kept alive, in good health and without symptoms of insufficiency for four months by oral administration of saline solution containing Ca, Mg, Na, and K chloride.—M. O. L.

Retention of vitamin C in Addison's disease (Das Verhalten des C-Vitamins bei Morbus Addisoni). Siwe, S., Klin. Wchnschr. **14:** 1311. 1935.

A patient with Addison's Disease, receiving a mixed diet but no added source of vitamin C, excreted a subnormal amount of ascorbic acid in the urine (compared with Harris and Ray's standards for the normal range). The oral administration of 0.6 gm. of ascorbic acid did not increase the excretion of this substance in the urine (contrast with normals of Harris and Ray), but a second dose of 0.6 gm. on the following day was followed by an increased urinary elimination of ascorbic acid. Third and fourth doses of 0.6 gm., administered before the urinary excretion had fallen to the control level, produced step-like increases in the excretory rate. In 41 hours, however, the rate of excretion fell to the control levels, in contrast to the more leisurely fall in the normals reported by Harris and Ray. The results are interpreted as indicating a relative deficiency of vitamin C in the tissues of this patient.—M. H. F.

Sodium chloride therapy in Addison's disease (Kochsalztherapie bei Morbus Addisoni). Siwe, S., Klin. Wchnschr. **14:** 1359. 1935.

In a precomatose patient with Addison's disease, whose blood sodium was definitely subnormal, it was found impossible to administer sodium chloride by mouth because such procedure provoked vomiting. After successful treatment with cortical extract had restored the patient's health, and the blood sodium to normal level, attempts to use the salt orally were frustrated by abdominal cramps and vomiting within 60 minutes after each dose, regardless of the fact that a number of vehicles were tried so that the patient was unaware that the salt was being administered. A severe grippe infection brought an another crisis in this patient some months later. The blood chemistry during this crisis showed no deviation from the normal, the blood sodium and the N.P.N. remaining at normal levels. From such findings, and from a perusal of the data in the literature, the author concludes that severe adrenal deficiency may exist without a decrease in the blood sodium or N.P.N., and that therefore neither the blood sodium nor the N.P.N. level can be used as a reliable criterion of early adrenal insufficiency, or as a guide for dosage of cortical extract.—M. H. F.

Effect of epinephrine on the sympathectomized human extremity; an additional cause of failure of operations for Raynaud's disease. Smithwick, R. H., N. E. Freeman and J. C. White, Arch. Surg. **29:** 759. 1934.

The fact that in animals structures innervated by the sympathetic nervous system become sensitized to circulating epinephrine following sympathectomy has been shown by Elliott as well as by Rosenblueth and Cannon. Evidence is presented in this report that human blood vessels become sensitized to epinephrine in the same manner following resection of sympathetic ganglions. Tests in a series of 9 cases in man following complete sympathetic denervation of extremities have shown marked vasospasm in the presence of minute quantities of epinephrine in the circulating blood. Thus the intravenous infusion of a 1:250,000 solution of epinephrine, an amount which causes little change in the normal extremity, is sufficient to lower the surface temperature of the denervated side as much as 15 F. Similar changes take place

when the patient's suprarenal glands are stimulated to secrete epinephrine by insulin hypoglycemia. Identical vasospastic phenomena, which occur in sympathectomized cats and rabbits in insulin hypoglycemia, are abolished by suprarenal denervation. This hypersensitization of the arteries to epinephrine takes place only on degeneration of the vasomotor nerves. It is not present after procaine hydrochloride block or during the first week after operation. The authors have found that it takes from 7 to 8 days for sensitization to appear. We believe that this hypersensitivity to the circulating hormone, epinephrine, which develops in sympathectomized extremities, constitutes a hitherto unrecognized but important source of unsatisfactory results in Raynaud's disease.—Authors' Summary.

The present status of the diagnosis and treatment of Addison's disease. Snell, A. M., M. Clin. North America **19:** 383. 1935.

About 80% of cases of Addison's disease are associated with tuberculosis of the suprarenal glands. The tuberculous process appears to begin in the medulla and to extend peripherally, thus cutting off fragments of the suprarenal cortex, which may be preserved to form the so-called cortical adenomas. These may, in the aggregate, amount to about 10% of the weight of a single suprarenal gland and are probably capable of some functional activity. In about 20% of cases atrophy of the suprarenal gland is responsible for the disease. Rarely carcinoma, gumma, hemorrhage, infarction, or chronic inflammatory changes with fibrosis may be the responsible pathological lesions. The incidence of Addison's disease in this country is about 300 to 400 cases annually, the death rate being 0.4 per 100,000 of the population. It is most apt to occur in the fourth or fifth decade of life, but cases have been seen in children. Males are affected about twice as often as females. The disease presents two characteristic sets of symptoms and signs: those of the stage of chronicity and those of crisis. The principal symptoms of the former are slight asthenia, hypotension, pigmentation of the skin, and occasionally evidences of hypoglycemia. The symptoms of crisis may develop at any time during the course of the chronic stage and are comparable to the symptoms observed in the bilaterally adrenalectomized animal. Outstanding are anorexia, nausea, vomiting, diarrhea and circulatory collapse, restlessness, delirium and coma, in association with significant changes in bodily chemistry. The association of pigmentation of the skin with demonstrable tuberculosis elsewhere in the body is of diagnostic significance; the presence of calcification within the suprarenal gland may be demonstrated by x-rays in about 25% of cases. By withdrawing salt from the diet of patients suspected of suffering with Addison's disease, it is possible to produce symptoms of crisis. This provocative test is only to be employed in a hospital. Diagnosis in the absence of pigmentation is a difficult matter, especially if the patient is dark complexioned. In treatment, the first indication is the maintenance of a sufficiency of sodium salts and fluids; the second is to supply the missing cortical hormone. Individualization in the management of these patients is important. A percentage respond to the administration of sodium chloride in large doses with little or no hormone treatment. Most patients require the combination of salt and hormone. Still another group respond poorly to treatment, suggesting the possibility of involvement of other glands, perhaps the anterior pituitary. The author cites a case in which addition of anterior pituitary treatment improved the patient's condition after the disease remained refractory to the specific therapeusis. Generally, the results of treatment are highly gratifying.—I. B.

Extraction studies on the adrenal cortical hormone. I. Methods of preparation. Pfiffner, J. J., H. M. Vars and A. R. Taylor, J. Biol. Chem. **106:** 625. 1934.

A quantitative determination of the amount of cortical hormone in beef adrenal glands extracted by various methods is given by assay on the adrenalectomized dog. The maximum yield is about 2500 dog units per kilo. Whole glands give as good yields as dissected cortex. When frozen there is a 25% loss in activity in the glands in six months. Neutral alcohol or acetone will extract all of the hormone. Glands dessicated at low temperature yield only one-tenth as much hormone as fresh glands. Benzene extraction removes hormone readily from 20% alcoholic solution. Permutit removes epinephrine and other toxic amines and causes a four-fold concentration of hormone. The fraction remaining after permutit can be kept in 95% alcohol for 2½ years at 6°C without loss of activity. Washing benzene soluble material with 4% $NaHCO_3$ to remove epinephrine causes loss of major portion of hormone.—J. M. L.

Differential depression of vasomotor mechanisms by adrenin. Wyman, L. C., and C. tum Suden, Am. J. Physiol. **113:** 271. 1935.

Pressor response to a small dose of adrenalin in 5 normal rats under urethane anesthesia was reversed by ergotamine, increased by cocaine and unaltered by atropine or physostigmine.

In 5 bilaterally adrenalectomized rats with transplanted cortical tissue, the depressor response to adrenalin was increased in two rats by ergotamine, reversed by cocaine and unaltered by atropine and physostigmine; while in the 3 others subcutaneous injections of adrenalin reversed these responses to small doses of adrenalin. They concluded that in the normal rat adrenergic dilator mechanisms were present but marked by depression from a secretion of adrenin. The same depressions of dilator mechanisms were demonstrated in one cat. In 3 guinea pigs and 3 rabbits ergotamine eliminated but did not reverse the pressor response to adrenalin. The differential depression of vasomotor mechanisms was suggested as significant in homeostasis.—A. J. Derbyshire.

Clinical aspects and morbid anatomy of pluriglandular sclerosis (Zur Kenntniss der Klinik und pathologischen Anatomie der multiplen Blutdruesensklerose). Boller, R., and A. Goedel. Wien. Arch f. inn. Med. 27: 41. 1935.

A clinical and morbid anatomical study of six cases is given with a discussion of the problems of "pluriglandular insufficiency." The authors consider Falta's terminology of pluriglandular sclerosis preferable to designate endocrine cachexia dependent upon changes of the main endocrine glands (pituitary, thyroid, adrenals and gonads) that are identical in nature and contemporary as to origin. It is often impossible to distinguish clinically between this disease entity and severe pituitary insufficiency (Simmonds' cachexia) accompanied by the customary secondary involvement of the other endocrine organs. The final differential diagnosis frequently remains with the pathologist who ascertains in pluriglandular sclerosis atrophy and sclerosis of the gland of a uniform type and apparently of similar age. These changes most probably represent sequels of some non-specific infectious process. This type is illustrated with one case history; a second case demonstrates the rare variety of tuberculous pluriglandular sclerosis. For the discussion of differential diagnostic points four more cases are presented, including one case of Simmonds' and Addison's disease respectively, a combination of myxedema and diabetes and one of combined adrenal and ovarian atrophy.—M. A. Goldzieher.

Endocrine studies in infants and children. I. Methods of procedure and diagnostic criteria. Gordon, M. B., J. Pediat. 5: 659. 1934.

The author presents forms of methods of procedure in the diagnosis of endocrine conditions in children. Diagnostic criteria are discussed and two illustrative cases are presented in which the methods described are employed.—M. B. G.

An experimental contribution to the study of the interrelations between the hypophysis, the thyroid glands and the testes (Contributo sperimentale a lo studio delle correlazioni fra ipofisi, tiroide, testicoli.) Pighini, G., Endocrinol. e pat. costit. 9: 230. 1934.

Eighteen adult male guinea pigs of equal body weight (550-600 grams) were selected. Six were castrated, 6 deprived of the thyroid glands and 6 were kept as controls. Two months after the operation 3 animals from each of the 3 groups received daily 0.5 cc. of a phenolated emulsion of pituitary gland which contained a strong dose of the sex-stimulating as well as thyrotropic component of the anterior lobe of the pituitary gland. Fifteen days later the animals were killed and the various organs (pituitary and thyroid glands, testes, seminal vesicles, prostate, penis) were histologically studied. The intact as well as the castrated animals which were treated with the pituitary extract showed evidences of pituitary hyperfunction—which was somewhat less pronounced in the animals deprived of the testes—while the castrated animals to which pituitary extract was not administered showed pituitary hypofunction. This shows that the pituitary gland reacts to the anterior pituitary hormone even in the absence of the testes. Also the thyroid glands were in a state of hyperfunction in the intact and in the castrated animals. The thyroidectomized animals did not show any histological change of the pituitary gland under anterior pituitary treatment while the histological picture of the testes was not clear-cut enough to draw conclusions.—A. Angyal.

The embryology of the thyroid, parathyroid and thymus of the Pacific pallid bat (Antrozous pacificus Merriam). Selle, R. M., Am. J. Anat. 56: 161. 1935.

Serial, transverse sections of 28 bat embryos ranging from 1 to 18 mm. in length, as well as serial sections of the tracheae and adjacent thyroids, parathyroids and thymus glands, of a full term fetus, of an immature male, and of 6 adult bats were studied. Models

were constructed of the developing pharynx and its derivatives. In this species of bat, four pairs of pharyngeal pouches develop. The single pair of thymus glands develop from the entire entodermal lining of the third pair of pouches, the single pair of parathyroid glands develop from a pair of epithelial masses formed at the distal and caudal ends of the fourth pair of pouches. The thyroid gland develops from a single, median anlage. This investigation has added another order of mammals (Chiroptera) to the list of those for which the development of the pharyngeal derivatives has been described, and shows that the development of these structures is less complicated than that recorded for other species.—Author's Abst.

Constitutional anatomical peculiarities of organs from the standpoint of postnatal development (Beiträge zur Kenntnis konstitutions—anatomischer Besonderheiten der Organe [vom Standpunkte der postnatalen Entwicklung]). Stefko, W. H., Ztschr. f. Konstitutionslehre **18:** 287. 1934.

The developmental history of the thyroid gland is reviewed. Five stages are recognized. The importance of these in studies on structural deviation is stressed. The thyroid of adult Mongolians (from a goiter-free region) resembles the immature thyroid gland of Europeans. Evidence of colloid formation from desquamated cells is also indicative of an infantile condition. The postnatal development of the suprarenal glands is detailed under 6 different stages, largely based on the thickness of the different layers of the cortex and the size of the medulla. The suprarenal glands of adult Mongolians is characterized by a failure of the cortex to reach complete differentiation and by its poor demarcation from the medulla. The large number of sympathetic ganglion cells in the medulla also indicates an infantile condition, as compared with the suprarenal glands of Europeans.—A. T. R.

Is osteogenesis imperfecta an endocrine disease (Ist die Osteogenesis imperfecta ein endokrines Leiden)? Ther, L., Virchows Arch. f. path. Anat. **395:** 57. 1935.

A gross and microscopic study of the endocrine organs from a case of osteogenesis imperfecta revealed no demonstrable lesions. The author therefore concludes it is not an endocrine disease.—R. L. Z.

The epilepsies associated with endocrine disorders. Turner, H. H., J. Oklahoma M. A. **27:** 389. 1934.

The author presents a brief résumé of the case histories and studies of 14 of his patients in whom convulsive states were associated with various endocrine disorders. All were relieved by endocrine therapy. Four of the reported cases were ascribed to parthyroid dysfunction; 3 hyperinsulinism or other hypoglycemic conditions; 3 adrenal; and 4 pituitary. The author demonstrates the importance of endocrine dyscrasias as an etiological factor in the causation of epilepsy. Calcium metabolism is mainly regulated by the parathyroids. One of the prominent actions of calcium is upon the excitability of the nervous and muscular systems. This is evidenced in parathyroid tetany by marked nerve and muscular hyperirritability, due to a lowered calcium balance. That many so-called epileptics are amenable to treatment is becoming more to be appreciated by the profession. The author contends that every sufferer from this symptom complex is entitled to, and should have, a thorough diagnostic survey, including a very careful neuropsychiatric and endocrine examination—the latter because there is evidence that hypofunction of the insular tissue, the adrenals and the hypophysis enhance cerebral irritability. Many epileptics become morose, depressed, and hypochondriacal because they have been wrongly impressed, by members of their family and by their physician, of the hopelessness of their affliction. Much can be done for these people by convincing them that they are not doomed, urging them to act and do as their fellow-men and attempting to lead them away from the egocentric fixation which they have acquired. Many can be relieved by endocrine therapy.—Author's Abst. (abbreviated).

Influence of the method used on the results produced in experimental cryptorchidism in rats and guinea pigs (Influence de la technique opératoire sur les resultats de la cryptorchidie expérimentale chez le rat et chez le cobaye). Brouha, L., and L. Desclin, Compt. rend. Soc. de biol. **117:** 67. 1934.

If the testes are sutured to the anterior abdominal wall, degeneration of the tubular epithelium is slight. If they are sutured to the posterior wall or simply forced inside the rings there is extensive tubular degeneration. Both rats and guinea pigs react in the same way.—J. C. D.

Studies on the mobilization of the symphysis pubis in the immature guinea pig (Recherches sur la mobilisation de la symphyse pubienne chez le cobaye impubère). Brouha, L., Compt. rend. Soc. de biol. **113:** 406. 1934.

Guinea pigs received enough rat units of folliculin to produce oestrus and then 1 milligram of corpus luteum extract free from progestin. Mobilization of the symphysis followed. If progestin was used after the folliculin there was no influence on the symphysis. This indicates two separate hormones from the corpus luteum.—J. C. D.

The effect of oestrin injections upon the developing ova of mice and rabbits. Burdick, H. O., and G. Pincus, Am. J. Physiol. **111:** 201. 1935.

Daily injections of oestrin begun on the day of mating ordinarily resulted in the retention of ova in the Fallopian tubes in both mice (15 cases—5 R.U. per day) the rabbits (7 cases—100-150 R.U. per day). All ova, whether they descended into the uterus or remained in the tubes, showed definite signs of degeneration by the fourth day after copulation. These observations indicate, therefore, that the prevention of pregnancy resulting from oestrin injections during the early preimplantation period may be due, a) to the degeneration of the ova long before they are capable of implantation and, b) secondarily to the prevention of uterine entry of the blastocysts. The retention of the ova in the tubes is probably due to a closure of the tubo-uterine junction as a result of oestrin action, although an inhibition of the normal ciliary mechanism is not excluded.—R. G. H.

Adhesion of fetal membranes and intersexuality in swine (Verwachsung der Fruchtsäcke und Intersexualität beim Hausschwein). Cohrs, P., Ztschr. f. Anat. u. Entwcklngsgesch. **102:** 584. 1934.

A description of 4 cases in domestic pigs in which the embryonic membranes had fused as has been observed more often in cows and small ruminants. Types of fusion and degrees of vascular anastomosis are discussed with their bearing on the influence one twin might have on the other.—A. T. R.

Studies on the amount of oestrus stimulating material in normal and ectopic testes (Recherches sur la teneur en substance oestrogène des testicules normaux et ectopiques). Courrier, R., Compt. rend. Soc. de biol. **117:** 1117. 1934.

Castrated female rats were used for testing the folliculin-like material extracted from the testis. None could be detected in bull testis. There was a definite amount in horse testis and more in the testes of pigs. The amount in undescended testes of pigs was about the same as that found in normally descended ones.—J. C. D.

Duration of estrus in ovariectomized and adrenal-ovariectomized rats before and after theelin. Emery, F. E., and E. L. Schwabe, Proc. Soc. Exper. Biol. & Med. **32:** 910. 1935.

Total adrenalectomy in rats has little or no effect on (a) the estrus cycles that occur after ovariectomy; (b) reducing the minimal dose of theelin; (c) the duration of estrus following 1 to 10 R.U. of theelin; (d) the tendency for estrus to reoccur after a positive reaction to theelin; and (e) reducing the dose of theelin needed to produce estrus in ovariectomized rats of long standing.—Authors' Summary.

Estrogenic treatment of gonorrheal vaginitis. Goldberg, L. E., C. L. Minier and E. L. Smith, J. Pediat. **7:** 401. 1935.

Amniotin, either by oral or hypodermic administration or by a combination of both methods, was given to 17 girls suffering from vaginitis. The ages ranged from 2 to 10 years of age. One girl had a non-specific infection while the others had gonorrheal infection. The disease recurred in the girl with the non-specific vaginitis while a cure was obtained in 14 of the 16 patients with specific vaginitis.—M. B. G.

Cytology of the mammary gland of the albino rat. Experimentally induced conditions. Jeffers, Katharine R., Am. J. Anat. **56:** 279. 1935.

The cytology of the mammary gland of the albino rat was studied in animals after parturition not followed by nursing a litter, after cesarean section, after hypophysectomy, after the injection of various extracts, in pseudo-pregnant animals, and in animals whose

uteri were distended with paraffin. The changes occurring in the cells are compared to those taking place during normal pregnancy, lactation and involution, and are correlated with endocrine function.—Author's Abst.

The treatment of amenorrhea with large doses of ovarian hormones (Die Behandlung der Amenorroë mit hohen Dosen der Ovarialhormone). Kaufmann, C., Klin. Wchnschr. **12:** 1557. 1933.

The author has repeatedly been able to obtain menstrual bleeding in castrated women by the administration of large doses of follicular hormone of the ovary. In one case a 28 year old castrated woman was given (hypodermically) 1,100,000 mouse units of progynon. After the last injection she manifested a definitely bloody discharge, while the curettage showed the presence of a glandular hyperplasia of the endometrium. However, in order to obtained a proliferative type of endometrium, in addition to the follicular hormone, injections of luteo-hormone were employed. Ampules of progynon-B containing 50,000 mouse units per cc. given twice a week were used in the investigation. The treatment was continued for 3 weeks in cases of primary amenorrhea and then corpus luteum hormone was administered for 5 days in doses of 25 and 50 rabbit units in this period. Five cases of primary amenorrhea with marked hypoplasia of the genitalia were treated with these hormones. Since the patients were young girls curettage was not performed on them. In two of the patients a distinct growth of the uterus could be determined by palpation. In two patients with primary amenorrhea and well developed genitalia treated with 200,000 mouse units of progynon and 50 rabbit units of luteo-hormone, menstruation began 2 days after the last injection of luteal hormone. The curettings showed a proliferative type of endometrium. The combined treatment in 3 cases of secondary amenorrhea with marked hypoplasia of the uterus resulted during the course of treatment in regular menstrual bleeding. However, spontaneous regulation of the menstrual cycle after cessation of treatment did not occur. In 2 other cases of secondary amenorrhea, repeated combined treatment resulted in several spontaneous periods. In several cases the use of progynon alone resulted in bleeding which microscopically was proved to be true menstruation with destruction of a pregravid endometrium. In 3 cases after the employment of progynon alone, regular menstruation occurred. —L. Goldstein.

The effect of Treponema pallidum on the metabolism of rabbit testis. Kraft, Ruth M., C. S. Robinson and S. Harris, Am. J. Syph. & Neurol. **19:** 354. 1935.

A study of the gaseous exchange in rabbit testicular tissue from 23 normal and 71 rabbits infected with Treponema pallida was made. The methods of Warburg and of Dickens and Simer were followed, using as a medium either serum or Ringer's solution buffered by bicarbonate or phosphate. The same qualitative relationships existed in the infected tissue as were observed in the normal tissue. The metabolic rate was decidedly reduced in the infected tissue, but the type of metabolism was not changed.—Ruth M. Kraft.

Action of folliculin on the fecundity of male mice (Action de la folliculine sur la fécondité de la souris mâle). Lacassagne, A., Compt. rend. Soc. de biol. **115:** 469. 1934.

The testes of male mice which received varying doses of folliculin did not atrophy. The only difference between normal males and males treated with folliculin was that the latter were rendered sterile by the injections.—J. C. D.

A note on the mechanism causing the disturbances in the ovary and oestrus cycle following a reduction in the amount of ovarian tissue (Sur le mécanisme intime des troubles ovariens et du cycle sexuel consécutifs à la réduction de la masse ovarienne). Lipschütz, A., and E. Vinals, Compt. rend. Soc. de biol. **116:** 1362. 1934.

In 7 guinea pigs 1 ovary was removed and the other broken into small fragments. Disturbances of the cycle and enlargement of the mammary glands followed. In 5 pregnant animals similar treatment was followed by abortion in 3. In 6 guinea pigs 1 ovary was fragmented and the other left intact. These animals showed no alterations in their cycle. In the cases where disturbances followed reduction of ovarian tissue there remained small bits of ovary with follicles in them. It would seem, therefore, that it is necessary to have a single mass of ovarian tissue of a definite minimum size in order to have it exert the normal restraining effect on the activity of the hypophysis.—J. C. D.

Prolonged action of urine of pregnant women and so-called extracts of the anterior hypophysis of urinary origin followed by loss of receptivity of the rabbit's ovary to these hormonal actions (Actions prolongées d'urines de femme enceinte et d'extraits dits antehypophysaires d'origine urinaire suivi de perte de receptivité de l'ovaire de lapin à ces actions hormonales). Moricard, R. Gynéc. et Obst. 31: 2. 1935.

The prolonged injection of urine and extracts of urine of pregnant women induced no characteristic cytological changes in the hypophysis but did affect the ovary. The author used 16 experimental animals and 2 controls. The injections were from 8 days to 5 months. There were 5 to 80 injections. The amount injected was from 15 cc. to 400 cc. containing 45 to 930 mouse units. Effect of urine and of extracts were equivalent. Injections for a short period were followed by increased follicular maturation. Later the follicles and corpora lutea diminished while the interstitial tissue increased. After 5 months the ovary was macroscopically normal but smaller than the ovary of an adult virgin rabbit.—J. P .P.

Glandular physiology and therapy; therapeutic use of estrogenic substances. Novak, E., J. A. M. A. 104: 1815. 1935. Abst., A. M. A.

The author states that estrogenic therapy is of little value in the treatment of endocrinopathic amenorrhea, although in many of these it is often resorted to because of the lack of any other treatment which is any more rational or any more effective. The weight of evidence indicates that in the treatment of menopausal symptoms, proper allowance being made for difficulties in the interpretation of the therapeutic results, the estrogenic substances are of real though variable value. The treatment of gonorrheal vulvovaginitis in children by means of injections of estrogenic substance is a promising method, though much more experience with it is necessary before worth while conclusions can be drawn as to the rapidity and permanence of the bacteriologic cure, and the possible harmful by-effects of the method. At present it seems that the treatment of hemophilia with estrogenic preparations will not live up to early expectations, though here again further experience with the plan must be awaited. While estrogenic therapy is at times employed for various other indications, as enumerated in the paper, the rationale is usually poorly defined and the results are ordinarily disappointing.

The reintegrative action of the anterior pituitary and of the ovarian extracts upon the experimental changes of the nasal mucous membranes and nasal bones produced by castration (L'azione reintegrative degli estratti anteipofisari ed ovarici sulle alterzione sperimentali della mucosa ed ossa nasali da castrazione.) Pighini, G., and C. F. Porta, Valsalva 10: 140. 1934.

There is a definite functional relationship between the gonads and the tissues of the nose. Borghesan has found previously that the bony structures and the mucous membranes of the nose undergo typical changes in the castrated young rabbits. The following experiments were conducted in order to show whether the change, due to castration, could be influenced by the action of sex-stimulating hormones. Twelve male and 12 female rabbits of the age of 10-12 months and of the body weight of 2200-2800 grams were used. Two males and 2 females served as controls. The others were castrated and 40 days later some of them received for 20 to 25 days daily 1 cc. liquor folliculi. Others received for the same length of time daily 1 cc. anterior pituitary extract. The histological picture of the nasal tissues in the castrated and non-treated animals confirmed the reports of Borghesan; softening and partial connective substitution in the nasal bones, atrophy of the mucous membranes and diffuse hyperaemia. These changes in the animals treated with liquor folliculi were much less noticeable and regenerative processes were clearly evident. The anterior pituitary extract had an influence similar to, but much more radical than the follicular extract.—A. Angyal.

The sex hormones of the female (Les hormones sexuelles femelles). J. A. Schockaert, Rev. med. de Louvain, Nos. 18 and 19, 1933; Nos. 6, 12, 13, 15, and 17, 1934.

This article is devoted to a résumé of the experimental and clinical findings on the hormones related to the female sex organs. Four hormones—the follicle stimulating and luteinizing principles of the anterior pituitary, and the estrogenic and corpus luteum hormones of the ovary are considered with regard to their nature, action, assay, and interrelations. The interactions of these several hormones in the growth and function of the female sex organs during infancy, puberty, pregnancy, and the menopause are considered.

The pathological conditions resulting from a deficiency or excess of each of the 4 hormones are dealt with in some detail. Emphasis is accorded the complications arising in the case of the pituitary hormones, due to the usual involvement of other hypophyseal functions. Means are presented for differentiating between primary and secondary amenorrhea, and between amenorrhea due to hypo- and hyper-folliculinism. Considerable attention is given to the possible involvement of a "sex center" in the floor of the third ventricle, and to the diencephalon in contributing to disorders of the sex organs. The value of the various pregnancy tests in diagnosing between normal and abnormal pregnancy, missed abortion, chorionepithelioma, etc. is reviewed in detail. Finally, methods of hormonal therapy in correcting the various disorders attributed to the over- or under-production of the sex hormones are suggested and discussed.—W. O. N.

Observations on reproduction in the chimpanzee. Schultz, A. H., and F. F. Snyder, Bull. Johns Hopkins Hosp. **57**: 193. 1935.

The chimpanzee, as regards its sex swelling, adolescent sterility, age of first menstruation and of first conception, was found to resemble the macaque more closely than it did man. As regards the duration of pregnancy, nature of pregnancy urine, and frequency of complications related to pregnancy, the chimpanzee stood closer to man. As regards the length of the menstrual cycle, man and the macaque were more closely alike than man and the chimpanzee.—A. G.

Comparatively low levels of oestrin in cases of chorioepithelioma and hydatidiform mole. Smith, G. V. S., and O. W. Smith, Proc. Soc. Exper. Biol. & Med. **32**: 847. 1935.

In 4 cases of chorioepithelioma the blood and urine contained little or no oestrin although large amounts of anterior pituitary-like gonadotropic hormone were present. Extraction of an hydatidiform mole also gave no oestrin.—M. O. L.

Contribution to the study of humoral antagonism in the rat ovary; the differing thresholds for the anti-luteinizing effects of folliculin (Contribution à l'étude de l'antagonisme humoral ovarien chez le rat. Seuils différentiels d'action anti-luteinique de la folliculine). Votquenne, M., Compt. rend. Soc. de biol. **117**: 1121. 1934.

Deciduomata may be produced in rats by placing a thread in the uterus during the first few days after birth of a litter when the uterus is under the influence of the corpus luteum. The antagonism between the corpus luteum and folliculin can be demonstrated if folliculin is injected during this period. An oestrus cycle is produced and the formation of deciduomata prevented if doses of 2 to 18 rat units are given.—J. C. D.

Effects of thyreotropic and adrenotropic hormones on hypophysectomized frog tadpoles. Atwell, W. J., Proc. Soc. Exper. Biol. & Med. **32**: 404. 1934.

The hypophysectomized frog tadpole is a striking test object for the action of pituitary hormones. This is especially true for the thyreotropic principle since the objective signs of metamorphosis are more readily observed than are changes in the basal metabolic rate. —Author's Summary.

An assay of three hormones present in anterior pituitaries of seven types of cattle classified for age, sex and stage of reproduction. Bates, R. W., O. Riddle and E. L. Lahr, Am. J. Physiol. **113**: 259. 1935.

A quantitative assay was made of several classes of cattle pituitaries to determine prolactin, the follicle-stimulating hormone (F. S. H.) and thyreotropic content. As revealed by testing in the immature male dove these principles were found in late-term embryos, adults of both sexes, castrate adult males and in pregnant cows. It was possible to run these assays simultaneously; increased testis weight showing quantity of F. S. H., increased crop weight showing quantity of prolactin, and increased thyroid weight indicating the thyreotropic content. The immature male dove proves a suitable test animal, as had been previously shown, in that the response of these three organs is independent of the others. Thyreotropic hormone occurred in fairly equal quantity in all types of pituitary, although rather low values were obtained from the glands of embryos and adult steers. F.S.H. occurred in greatest amount in glands from cows in early pregnancy, and least in adult male castrates. Nearly equal amounts of prolactin were found in the anterior lobes of veal calves, adult steers, adult bulls and non-pregnant cows; larger amounts were present in cows

both in early and late pregnancy. The largest quantities occurred in the whole glands of 5-7 month embryos. The method of extraction of these principles is included.—H. O. Haterius.

Glucose tolerance and hypophyseal deficiency (Insuffisance hypophysaire et tolérance au glucose). Biasotti, A., Compt. rend. Soc. de biol. **117:** 54. 1934.

Fourteen dogs were hypophysectomized, 5 had the region of the tuber cinereum damaged and 10 were controls. The hypophysectomized animals failed to dispose of the glucose injected into the blood stream as fast as the controls. The response of the dogs with injured tuberal regions was only slightly slower than in the controls.—J. C. D.

Blood fat tolerance tests in malnutrition and obesity. Blotner, H., Arch. Int. Med. **55:** 121. 1935.

This paper presents a study of alimentary fat tolerance, using the plasma cholesterol as an indicator of the blood fat concentration, and as a test meal 500 cc. of 20% cream. In normal persons the plasma cholesterol level following the ingestion of such a test meal remains practically unchanged during an eight hour period. In thin persons the cholesterol level after such a test meal remains unchanged or even shows a decrease. Under insulin therapy, however, there is a significant rise in the cholesterol level after such a test meal. In obese persons there is a marked rise in the cholesterol level after such a test meal. The blood fat curve in obesity is similar to that obtained in malnourished people receiving insulin. In diabetes insipidus after such a test meal the cholesterol level increases markedly, and the resultant curve is similar to that obtained in obesity. Extract of pituitary administered intranasally modifies the plasma cholesterol curve obtained after a fat test meal in patients with obesity and diabetes insipidus. Extract of pituitary prevents the development of hyperlipemia which otherwise occurs. Insulin and extract of pituitary have effects on fats as well as on sugar metabolism that are of opposite nature, insulin causing a rise in the blood fat concentration after a fat meal and extract of pituitary a fall. In a group of 6 obese patients treated for short intervals with applications of extract of pituitary intranasally and without diet there has been an average weekly loss of from 1½ to 2 pounds. The greatest amount of weight lost under this form of therapy by any one patient has been 17 pounds in 10 weeks. Although these results are of some interest, no definite conclusions can be drawn regarding their significance. The possibilities of such a method of treating obesity will remain uncertain until further studies have been completed.—Author's Summary.

The influence of epinephrine on the blood sugar, lactic acid and inorganic phosphorus of completely hypophysectomized dogs. Chaikoff, I. L., F. L. Reichert, L. S. Read and M. E. Mathes, Am. J. Physiol. **113:** 306. 1935.

Hypophysectomized dogs (H) after complete recovery from operation (1-4.5 months) respond to subcutaneous epinephrine injections in a manner qualitatively similar to normal dogs (N), i.e. by a rise in blood sugar and lactic acid and a fall in inorganic P. The magnitude of the response is less by about 50% for the 3 blood constituents. Seven dogs in group H and 4 in group N were used. That the altered response to epinephrine is due directly to removal of the hypophysis is shown by the fact that 4 dogs (C), in which all operative procedures but actual removal of the hypophysis were performed, exhibited the same response to epinephrine as unoperated dogs (N). Explanations are discussed and no conclusions are drawn.—A. H. Hegnauer.

Pregnancy cells in rat pituitary; influence of lipoidal corpus luteum extract. Charipper, H. A., Proc. Soc. Exper. Biol. & Med. **32:** 402. 1934.

Evidence is offered that the essential factor for the production of pregnancy cells in the anterior pituitary of the rat is contained in the luteinized ovary and that this factor is not sex specific.—Author's Summary.

Basophil adenoma of the pituitary gland with renal changes. Close, H. G., Brit. M. J. No. 3868: 356. 1935.

A case of basophil adenoma of the pituitary is described in which the only symptom of the many forming Cushing's syndrome was a high blood pressure. The finding of malignant nephrosclerotic changes in the vessels of the kidneys is additional evidence in favour of the suggestion that the hypertension in cases of this pituitary lesion tends to be of the "malignant" type.—Author's Summary.

Concerning gonadotropic substances in mare serum. Cole, H. H., and G. H. Hart, Proc. Soc. Exper. Biol. & Med., **32:** 370. 1934.

Sera from non-pregnant mares and from mares 80 and 180 days pregnant were injected alone and in combination with pituitary synergist (made by method of Evans et al.) into immature female rats. Although 2 cc. of mid-pregnancy serum when given alone induced follicular development and luteinization and 10 cc. or even 60 cc. of non-pregnant serum produces no response, equivalent amounts of these two types of sera are equally effective in stimulating the ovaries when combined with pituitary synergist. Likewise, 0.04 cc. of early pregnancy serum is effective alone but when combined with pituitary synergist the response does not exceed that of non-pregnant serum plus synergist. Thus it is concluded that non-pregnant serum contains a single gonadotropic substance only effective in combination with pituitary synergist while pregnancy serum contains this substance plus another substance effective by itself.—Authors' Abst.

Adrenotropic hormone of anterior pituitary lobe. Collip, J. B., Evelyn M. Anderson and D. L. Thomson, Lancet **2:** 347. 1933, Abst., J. A. M. A. **101:** 1913.

Evidence is presented that the suprarenotropic principle as obtained in refined extracts is distinct from the thyrotropic principle. The hypophysectomized rat is a satisfactory test animal for detecting the presence of the suprarenotropic hormone. A series of tests on 125 rats which were completely hypophysectomized is reported: (1) loss of weight following hypophysectomy, (2) complete failure to grow, (3) atrophy of the left suprarenal removed for control examination before starting the injections and (4) absence of any fragments of the pituitary by careful inspection at the time of necropsy. In some of the animals the authors have had further evidence of the completeness of the hypophysectomy by determining the basal metabolic rates and by serial sections of the sella turcica. The animals used were hypophysectomized from 11 to 148 days previously. Before starting the injections, the left suprarenal was removed, weighed and sectioned for histologic examination. Removal of one suprerenal from the hypophysectomized rat is not followed by a compensatory hypertrophy of the other suprarenal, such as occurs in the normal animal. In six control animals from which the left suprarenal was removed from 8 to 23 days after hypophysectomy and the right suprarenal one week later, the right suprarenal in each case weighed either the same or less than the left suprarenal of the same animal and all showed marked atrophy of the cortex by histologic examination. In another group of 7 hypophysectomized animals receiving Witte's peptone (20 mg. daily) or extracts of anterior pituitary free from the suprarenotropic hormone, there was no evidence of any repair of the atrophy that is seen in the untreated hypophysectomized rat. Of 40 different extracts of the anterior pituitary lobe that have been tested, 34 were found to contain the suprarenotropic hormone. Eight of these extracts showed a fairly high degree of potency, causing an increase of from 50 to 300% in the weight of the right suprarenal over that of the left suprarenal, which had been removed previously as a control. Two of the extracts that have been assayed brought about repair of the suprarenal in the hypophysectomized rat when 0.025 cc. (0.125 mg. of total solids) was administered twice a day for 1 week. Four extracts with a high degree of potency showed no trace of the thyrotropic hormone. These extracts had no effect on the growth of the body. Extracts boiled for 30 minutes still showed suprarenotropic activity. Active extracts were found to be ineffective when administered orally. The authors obtained the purest extracts of the suprarenotropic principle from a fine flocculence, which separated out on concentrating a 75% acetone-soluble fraction at from pH 5 to 6. The latter was obtained as a by-product in the purification of the thyrotropic fraction in the filtrate of the original aqueous extract after the tricalcium phosphate adsorption treatment to remove growth hormone.

Hyperactivation of the neurohypophysis as the pathological basis of eclampsia and other hypertensive states. Cushing, H., Am. J. Path. **10:** 145. 1934.

Cushing found that the posterior lobes of pituitary glands were massively infiltrated by basophilic cells from patients who had died of pituitary basophilism and had had severe hypertension while living. Four patients who died in eclampsia or toxemia of pregnancy with hypertension had similar pituitary findings while 2 such patients showed no basophilic invasion of the posterior lobe. From patients who died of essential hypertension, Cushing found also a massive basophilic invasion of the posterior lobe of the pituitary. Even patients who have had hypertension associated with generalized arteriosclerosis, or malignant hypertension were found to have such basophilic invasions of the pars nervosa of the pituitary.

He believes that the pars intermedia is the source of supply of the basophilic cells that invade the posterior lobe and that the extent of the invasion is a measure of posterior lobe activity. He further believes this invasion may be the etiological relationship in hypertension states.—W. Freeman.

Carbohydrate metabolism; the effect of hypothalamic lesions and stimulation of the autonomic nervous system. Davis, L., D. Cleveland and W. R. Ingram, Arch. Neurol. & Psychiat. **33:** 592. 1935.

Hyperglycemia and glycosuria are not produced by pancreatectomy in cats which have a lesion of the hypophysis. Bilateral hypothalamic lesions, symmetrically placed, may be followed by pancreatectomy without the development of hyperglycemia and glycosuria. Such lesions must be situated in the tuber cinereum slightly rostrodorsolateral to the mamillary bodies at the level of the ventromedial hypothalamic nucleus. Stimulation to the superior cervical sympathetic ganglion and the stellate ganglion in cats produces marked hyperglycemia and glycosuria. These results are not obtained following section of the splanchnic nerves of bilateral, symmetrical lesions of the hypothalamus.—Authors' Summary.

A gastric lesion produced by an extract of the pituitary gland. Dodds, E. C., R. L. Noble, and E. R. Smith, Lancet **2:** 918. 1934.

An extract of the posterior lobe of the pituitary gland has been prepared which on subcutaneous injection into rabbits, mice, rats and guinea pigs produced in 12 to 15 hours an intense haemorrhagic gastritis. The lesion was confined strictly to the acid-secreting area of the stomach and was produced in every case in 40 experiments on rabbits. Similarly prepared extracts of many control tissues gave completely negative results even in very large doses. The active substance was shown to be present in standard B.P. pituitary extract, which was also active when given by mouth. The oxytocic fraction of Kamm gave negative results but the pressor fraction gave the typical response. The mode of action and possible physiological rôle together with further purification of this substance is under investigation. Such large doses were necessary that the amount of the gastrotoxic factor contained in ordinary clinical doses of pituitary extract would appear to be negligible.—R. L. Noble.

Anterior hypophysis and thyroid gland. The influence of thyreotropic substance upon liver glycogen and blood ketone bodies (Hypophysenvorderlappen und Schilddrüse. Der Einflus der thyreotropen Substanz auf Leberglykogen und Blutketonkörper). Eitel, H., G. Löhr and A. Loeser, Arch. f. exper. Path. u. Pharmakol. **173:** 205. 1933.

Twelve white male rats (140-170 gm.) served as controls to determine the normal glycogen content of the liver and ketone body content of the blood. These determinations were made on well-fed animals without previous preparation by starvation or by feeding known quantities of carbohydrate; and similar determinations were carried out on 9 white male rats after thyroidectomy. The authors state that large doses of thyreotropic substance (anterior pituitary extract) caused a marked decrease in the glycogen content of the liver and an increase of the ketone body content of the blood in previously normal animals. They conclude that these effects could not be elicited in 9 thyroidectomized rats under the same experimental conditions. The glycogen content of the livers from the 9 thyroidectomized animals which controlled the latter experiment and on which this conclusion is based, ranged from 0.7 to 6.1%.—H. B. F.

Gonadotropic hormone (prolan) in relation to carcinoma of the cervix. Halstead, J. A., New England J. Med. **213:** 803. 1935.

The excretion of gonadotropic hormone was estimated, by Zondek's method in the urines of 15 women with carcinoma of the cervix. Positive results were obtained in 4 cases. The incidence of positive findings 26.6%, though greater than occurring in normal women is not sufficient to be of diagnostic significance.—J. M. L.

Diabetogenic action of anterior pituitary substance under various physiological conditions (Action de la substance diabétogène ante-hypophysaire dans diverses conditions physiologiques). Houssay, B. A., A. Biasotti and C. T. Rietti, Compt. rend. Soc. de biol. **115:** 323. 1934.

The authors have carried out numerous experiments on dogs in which they observed the usual increase of blood sugar following administration of anterior hypophyseal extract

even after removal of the pancreas, hypophysis, gonads, adrenal medulla, thyroid, etc.—J. C. D.

Pituitary hormones and the blood sugar level. Hrubetz, M. Caroline, Proc. Soc. Exper. Biol. & Med. **32:** 842. 1935.

No changes from the normal in the blood sugar level were found in 10 rats injected with antuitrin S, 6 with antuitrin G, 5 with theelin, and 10 with anterior pituitary extract, Squibb.—M. O. L.

Hormonal investigations in Cushing's disease (Über Hormonuntersuchung bei Morbus Cushing). Jores, A., Klin. Wchnschr. **14:** 1348. 1935.

Hormonal investigations were conducted on a multipara of 35 who presented a typical picture of Cushing's disease, but who was menstruating regularly at the time. The Zondek-Aschheim reaction was negative in both blood and urine. Tests for the melanophore principle in the urine by the author's method (technique not given; no references given) indicated somewhat elevated values, although the Zondek-Krohn method (on "Ellritze") and the isolated frog skin method failed to reveal any increase of this principle. The injection of a protein free plasma preparation from this patient produced an adrenotropic effect greater than normal when injected into immature mice (author's method; to be published). The degree of this adrenotropic effect was comparable to that evoked by a solution of posterior pituitary powder containing 0.2 gm. per cc. On the basis of such findings the author concludes that there is an increased elimination of melanophore and adrenotropic factors in Cushing's disease, and in the light of such conclusions, discusses the hormonal relations of this disease to other pituitary disorders.—M. H. F.

The quantitative determination of small amounts of gonadotropic material. Katzman, P. A., and E. A. Doisy, J. Biol. Chem. **106:** 125. 1934.

The authors give details for three methods of concentrating the gonadotropic material in normal urine. In the first 2 the material is precipitated by tungstic acid and liberated by barium hydroxide or by brucine. In the third method the material is precipitated by benzoic acid, extracted with sodium hydroxide and precipitated by acetone. This makes possible a 200-fold concentration and gives a less toxic product than the tungstate method. The material is assayed by the vaginal-canalization and the estrus-production method in immature rats or mice. In normal adult males from 0 to 19 mouse units of gonadotropic material per 24 hours were found. In girls between the ages of 4 years and puberty little or no gonadotropic material was found, but at puberty an increased excretion occurred. A maximum excretion of gonadotropic material occurred in the urine of normal women about the time of ovulation or during menstruation. At other times the values fall to 1 rat unit or less. Up to 75 rat units per 24 hours may be found in urine of ovariectomized women. This material is different from that obtained from pregnancy urine.—J. M. L.

The anterior pituitary, iodine and the thyroid gland. Loeser, A., and K. W. Thompson, Internat. Clin. **3:** 149. 1935.

This paper deals with the relatively neglected subject of the relationship of iodine to thyroid-pituitary conditions. Now that the fact is known that a primary mechanism for activation of the thyroid emanates from the pituitary, how does iodine produce thyroid stimulation or depression? Experimenting on rats, the authors find (1) iodine does not activate the thyroid in the absence of the pituitary gland; (2) in hypophysectomized rats the activating effect of thyreotropic hormone plus iodine is not greater than the effect of thyreotropic hormone alone; (3) large doses of iodine tolerated by normal rats are apt to be fatally toxic to hypophysectomized rats of the same size. It would appear that in response to the presence in the blood of ionic iodine, the hypophysis varies its production of thyreotropic hormone and thus in turn the output of the metabolic hormone of the thyroid is varied, a process which in itself may utilize and detoxify iodine.—I. B.

Observations upon hypophysectomized-depancreatized cats. Long, C. N. H., and F. D. W. Lukens. Proc. Soc. Exper. Biol. & Med. **32:** 326. 1934.

Observations were made on 2 cats in which total pancreatectomy was performed 2 weeks after hypophysectomy. They survived 35 and 85 days. No insulin was given at any

time. In 9 control animals the average survival after pancreatectomy was 4 days. In general the findings were comparable to those reported by Houssay and others in the dog. The ameliorating effect of hypophysectomy upon pancreatic diabetes has been demonstrated in other species of animal. A limited number of observations showed that these cats developed no ketonuria although the other characteristics of diabetes—glycosuria, polyuria and marked loss of weight, were present in varying degrees.—Authors' Abst.

Lactic acid in the blood of hypophysectomized dogs (Acide lactique sanguin des chiens hypophysoprives). Marenzi, A. D., Compt. rend. Soc. de biol. **117:** 53. 1934.

In 14 hypophysectomized animals the lactic acid did not differ from the normal as represented by 18 controls.—J. C. D.

The mineral substances in the plasma of hypophysectomized dogs (Substances minérales du plasma des chiens hypophysoprives). Marenzi, A. D., and Mlle R. Gerschman, Compt. rend. Soc. de biol. **117:** 56. 1934.

In 6 hypophysectomized dogs in good health, 12 to 297 days after operation, there was a 17% diminution of potassium and 11% of magnesium as compared to that of 9 to 22 controls. The other mineral elements did not vary abnormally.—J. C. D.

The site of the antidiuretic action of pituitary extract. Marshall, E. K., Jr., W. W. Burgess, and A. M. Harvey, Tr. A. Am. Physicians, **48:** 219. 1933.

In the many experiments in which pituitary extract has been injected into various mammals after the ingestion of water, it has been universally found that the water diuresis is almost completely inhibited for several hours, but the site of this antidiuretic action has been the subject of much experimentation and controversy. Two general theories have been proposed, that of an extrarenal action on the water exchange between the blood and tissues and that of a direct action on the kidney which cannot be accounted for by renal vascular changes. No convincing evidence to support the theory of an extrarenal action has been presented. In considering the site of the antidiuretic action of pituitary extract on the kidney, two possibilities must be borne in mind: (a) A decrease in the quantity of glomerular filtrate, and (b) a stimulation of water reabsorption by some part of the renal tubule. Using the xylose and sucrose method of measuring the glomerular filtrate, the authors have found no change in the quantity of glomerular filtrate as a result of the antidiuretic action of pituitary extract in the mammal. This eliminates the possibility of blood pressure changes in the glomerulas or a decreased permeability of the glomerular capsule explaining the action of this drug. The only explanation would appear to be a stimulation of water reabsorption by some part of the renal tubule. In tests of the action of pituitary extract on a water diuresis produced in various classes of animals, no antidiuretic action was observed in the fish or amphibian (frog). In the reptile (alligator) a marked antidiuretic effect was produced; further experiments showed that this is not due to increased water reabsorption by the tubule, but to a decrease in the amount of glomerular filtrate. In the bird (chicken) an antidiuretic effect was observed which, with small doses, was found to be due entirely to an increased water reabsorption by the tubule. Larger dosage produced, in addition, a decrease in glomerular filtrate. The conclusion appears justified that the antidiuretic action of pituitary extract in man is due to a stimulation of water reabsorption by the loop of Henle. The lack of any antidiuretic action in the fish or amphibian would argue against any extrarenal action of pituitary extract.—I. B.

The presence of ciliated cells in the anterior lobe of the hypophysis of the albino rat (Sur la présence de cellules ciliées dans le lobe anterieur de l'hypophyse du rat blanc). Martins, T., Compt. rend. Soc. de biol. **113:** 216. 1933.

In two rats and one marsupial (Didelphys aureta Wied) the authors saw tubes lined with ciliated cells. These are described.—J. C. D.

Studies on the influence of urine from acromegalics on the growth of young animals (Recherches sur l'action de l'urine des acromégaliques sur la croissance de jeunes animaux). Parhon, C. I., C. Parhon-Stefanescu and E. Tomorug, Compt. rend. Soc. de biol. **117:** 144. 1934.

Two rats, 5 guinea pigs, and a rabbit, with suitable controls, were injected with urine extract over a 30 day period. Excluding obviously pathological animals there was a more

rapid growth in those receiving acromegalic urine. This excess amounted, however, to only 6.3% by weight.—J. C. D.

Experimental diabetes insipidus; its relation to the anterior and posterior lobes of the hypophysis. Richter, C. P., Am. J. Physiol. **110:** 439. 1934.

The pituitary gland was totally removed in 34 rats; partially in 32. Total removal of the gland produced a temporary diabetes insipidus in 28 animals, a permanent diabetes in none. A permanent diabetes insipidus was found in 26 animals in which the posterior lobe was entirely removed but in which part of the anterior lobe remained. Six animals in which parts of the posterior lobe remained (probably still in connection with the stalk) showed no diabetes insipidus. The degree of the permanent diabetes insipidus seems to depend on the amount of the anterior lobe remaining, and whether or not this lobe is still in connection with the pituitary stalk. The present research regarding the importance of the anterior lobe in diabetes insipidus confirms the conclusion arrived at by von Hann in 1918 in a study of clinical material in man.—Author's Summary.

Effect of thyreotropic hormone preparation of the anterior lobe of the hypophysis on diuresis in the white mouse and on the blood pressure of the cat. Anterior-posterior lobe antagonism (Wirkung eines thyreotropen Hormonpräparates des Hypophysenvorderlappens auf die Diurese der weissen Mause und den Blutdruck der Katze. Antagonismus Vorderlappen-Hinterlappenhormone). Rothschild, F., H. Staub and K. Mezey, Arch. f. exper. Path. u. Pharmakol. **179:** 61. 1935.

A study of the diuretic and blood pressure effects of thyreotropic hormone of the anterior lobe as compared to those of the posterior lobe was made using 120 male white mice and 7 urethanized cats. By Gibbs' method of urine collection an initial antidiuresis of 2 hours duration was observed following intraperitoneal injection of thyreotropic hormone; diuresis followed in proportion to the dosage (20-100 M. E.). Posterior lobe extract quickly checked this. Qualitative and quantitative comparisons of blood pressure and diuretic actions of anterior lobe thyreotropic preparations and posterior lobe extracts led authors to believe the initial antidiuresis following thyreotropic injections to be due to posterior lobe activity. 1-2000 M. E. of thyreotropic hormone lowered blood pressure in cats. Authors believe this a specific action of the anterior lobe preparation and that a regulatory mechanism exists between anterior and posterior lobe for both blood pressure and diuresis.—R. Cleveland.

The gonadotropic hormone content of human anterior hypophyses (Gehalt des menschlichen Hypophysenvorderlappens an gonadotropen Hormonen). Schockaert, J. A., and H. Siebke. Zentralbl. f. Gynäk. **57:** 2774. 1933.

Pointing out that the evaluation of the hormone content of pituitary tissue by the implantation method is subject to many errors, the authors have adopted a simple extraction procedure. They weigh the gland, grind it with sterile sand and water, centrifuge and dilute the extract with sterile water to 20 cc. If necessary, further dilutions are made of this standard extract. Using this method, and employing infantile female mice, they have assayed pituitaries from 18 women, 18 men, and 3 newborn infants for both follicle-stimulating and luteinizing hormone. The hormone content of glands from newborns is negligible, and from women after childbirth or abortion is very low. Little difference in gonadotropic hormone content was observed between the two sexes. Female pituitaries contained up to 4000 m.u. of follicle-stimulating and 1500 m.u. of luteinizing hormone, while male glands assayed up to 3000 m.u. of follicle-stimulating and 1000 m.u. of luteinizing hormone. However, the average content for both sexes was approximately 1000 m.u. of follicle-stimulating and 400 m.u. of luteinizing hormone.—W. O. N.

Estimation of the thyreotropic activity of the anterior pituitary gland upon the basal metabolism (Auswertung der thyreotropen Wirkung des Hypophysenvorderlappens am Grundumsatz). Schoedel, W., Arch. f. exper. Path. u. Pharmakol. **173:** 314. 1933.

Metabolism studies on 26 normal untreated male guinea pigs weighing between 300 and 500 gm. showed that their average rate of oxygen consumption (138 determinations) was 6250 cc. 0_2 per sq. m. per hour $\pm 8.5\%$. Individual animals showed variations of $\pm 3\%$. Various doses of two extracts of the anterior hypophysis were injected into groups of 2 to 4 guinea pigs (total of 29 animals) to determine whether the maximum increases in the rate of metabolism paralleled the size of the dose. The latter consisted of 3 daily injec-

tions either of Schering's thyreotropic hormone (ThHS) in doses of 6.25 to 100 guinea pig units or of "acetone dry powder" (ATP) in doses of 100 to 400 mg. The maximum increases in the rate of oxygen consumption varied considerably in each test group of 2 to 4 animals. The minimum variation was 11% and the maximum, 31%. The 4 guinea pigs receiving 100 units of extract daily yielded smaller increases in metabolism than the four which were given 50 units daily. The maximum increases in the rate of metabolism of the four animals receiving 3 daily injections of 25 units ranged from 15 to 37%. On the basis of these results the author concludes that a quantitative relationship exists between the extent of change in the rate of metabolism and the dose of thyreotropic substance, when the latter lies between 10 and 40 units daily. The author includes a statement to the effect that the morphological changes in the thyroid gland run parallel with the estimation of the metabolic rate, but no data are recorded.—H. B. F.

Cachexia of pituitary origin and related conditions (Ueber hypophysäre Kackexie und verwandte Zustände). Steinitz, E., Deutsche med. Wchnsch. **58:** 1323. 1932.

Clinical observations were made on six patients. Cachexia of pituitary origin may be due to tumor, lues, thrombosis or embolism of the gland, but the classical cases in women are according to Reye due to an atrophy of the anterior lobe post partum. The author believes that it is not always an atrophy of the anterior lobe that is the cause of cachexia, but often a disturbance of the internal secretion alone, which is reparable. The disposition to the cachexia is present not only after pregnancy, but also after other physiological hyperactivity of the anterior lobe, as in puberty and after the menopause. Three groups of symptoms can be distinguished, senilism with senile skin and loss of hair on head and body, wasting with stubborn loss of appetite, sometimes trophic changes of the joints, and finally sexual disturbances with amenorrhoea. Patients treated with pituitary preparations by mouth, or with parenteral administration of Praephyson or other anterior lobe extracts alone lost only the trophic changes of the joints, but the loss of weight continued. In spite of the fact that the fasting blood sugar levels were below normal, the glucose tolerance test indicated a lack of insulin. The blood pressure in all cases was below normal. Treatment with 8 to 12 units of insulin per day and Ephetonin to elevate the blood pressure plus pituitary preparations per os gave very satisfactory clinical results. The patients regained weight up to 3½ kgn. per week, the blood pressure increased and the total constitution of the patients changed toward normal. X-ray examination of the chest showed the change from a paralitic thorax with the heart of drop-form in the height of the disease, to a normal chest and almost slightly dilated heart. The behavior of the patients changed from the irritated, uncooperative one in the disease to a normal one. Libido and mental ability appeared again.—R. J. Wagner.

Effect of hypothyroidism on antidiuretic action of pressor principle of posterior pituitary. Stern, Beatrice, and D. R. Gilligan, Proc. Soc. Exper. Biol. & Med. **32:** 843. 1935.

The antidiuretic effect of the pressor principle of posterior lobe pituitary (pitressin) injected subcutaneously is the same in patients with hypothyroidism as in subjects with normal thyroid function. The blood pressure and heart rate are not appreciably affected, either in normal subjects or in patients with hypothyroidism, by the subcutaneous injections of 0.10 and 0.15 cc. of pitressin.—Authors' Summary.

Adenomata of the pituitary, with special reference to pituitary basophilism of Cushing. Susman, W., Brit. J. Surg. **22:** 539. 1935.

In 260 cases of routine necropsy examinations, the hypophysis was removed and the infundibular region was examined. Of these glands 22 contained 23 tumors, and, of these, 20 were of a purely incidental character and had given rise to no symptoms. Of this last group 8 were of the basophilic type. The data show that incidental tumors occurred in 8% of the cases in this series when the gland was examined in the infundibular region alone and that the basophilic tumor was the commoner type present. The age and sex distribution were roughly comparable to those of the general series. It was also found that in 21 cases of the whole series the basophilic count was definitely high. These cases showed no unusual characteristics in relation to age, sex or disease groups. The common occurrence of pituitary adenomata (8%) and particularly those of the basophilic type (3%) would cast doubt on the association of a syndrome such as "pituitary basophilism" with the occurrence of such cells.—H. Freeman.

The effect of pituitary (posterior lobe) extract upon the urinary flow in non-anaesthetized dogs. Samaan, A., J. Physiol. **85:** 37. 1935.

The action of posterior pituitary extract on urinary flow was observed in 5 bitches trained to receive a stomach tube and remain quiet in a Parlor stand. Three of these had one kidney denervated and both ureters exteriorized. The other two had the exterior urethral opening exposed by perinectomy, one of the animals later had both kidneys denervated. After establishment of a uniform diuresis the water intake was adjusted to balance the renal and extrarenal output. The "anti-diuretic threshold dose" of extract given intravenously was fairly constant for any one animal, being quantitative for 20% variations in dose. Denervation of the kidney did not affect water diuresis and its inhibition by posterior pituitary extract. The diuretic effect described in anaesthetized animals was not observed. The extract did not diminish the flow of urine induced by ingestion of concentrated solutions of urea. —R. L. Z.

Experimental pituitary basophilism. Thompson, K. W., and H. Cushing, Proc. Roy. Soc., London, S. B. **115:** 88. 1934.

An extract of hypophyses of sheep containing active gonadotropic hormone was administered subcutaneously for 90 days to a pedigreed fox-terrier, female, whose littermate sister was used as a control through the experiment. They were 8 weeks old when the experiment was begun. From her sister, who during the 93 days of observation grew into a lanky, active animal, the injected puppy differed in the following ways: 1) She had become exceedingly adipose, slow in movement, and petulant in disposition, yet appeared in good health. 2) There was retardation of skeletal growth, delay in normal dentition, and extremely slow growth of hair. Serum calcium values were invariably higher than in the control, yet decalcification of bones could not be demonstrated. There was no hyperplasia of parathyroid glandules. 3) Compared with the control, she had a markedly subnormal basal metabolic rate and the thyroid proved to be colloidal in type with extremely flat epithelium. 4) The carbohydrate tolerance was diminished though there was no spontaneous glycosuria. The liver was heavily loaded with glycogen to a degree seen only in von Gierke's disease. 5) No nephro-vascular effects were evident; the blood pressure was subnormal. A striking pallor of the tissues and a secondary type of anemia were present. 6) Normal sexual development was much retarded in the injected puppy. A number of these changes strongly suggest experimental reproduction of certain features of the clinical syndrome of pituitary basophilism.—Authors' Abst.

Problems of the new-born. Bonar, B. E., J. Pediat. **5:** 230. 1934.

Diabetes in pregnancy is more common than formerly. Prior to the advent of insulin, the majority of diabetic women were sterile. While pregnancy aggravates diabetes, the maternal dangers are not much greater than for pregnancy uncomplicated by diabetes. Maternal deaths are usually due to coma and acidosis rather than to intercurrent infections. The fetal death rate has not kept pace with the great reduction in maternal mortality—it is still over 40%. Reduction can come only through the control of diabetes during pregnancy. There is a high percentage of birth injuries due to the tendency of the infants to be large. Correct management of the mother assures a better chance for life of both mother and infant.—M. B. G.

Blood sugar regulation in exercise. Dill, D. B., H. T. Edwards and S. Mead, Am. J. Physiol. **111:** 21. 1. 1935.

The finger blood sugar was determined during the course of treadmill work of widely varying intensities and also during recovery. The subjects of most of the experiments were two normal young men, one a trained distance runner. Easy work did not significantly affect blood sugar. Exercise involving 12-18 times the resting energy output caused increases during work of 10-66% followed usually by still higher levels in the first 10 minutes of recovery. Intense work, chiefly anaerobic, caused little change in the blood sugar during work and only small increases in recovery.—S. Mead.

Do the salivary glands have an internal secretory effect on the carbohydrate metabolism (Wirken dies Speicheldrüsen auf den Kohlehydratstoffwechsel inkretorisch ein)? Glaser, E. and I. Bannet, Klin. Wchnschr. **12:** 345. 1933.

The authors analyzed chemically a few kilograms of salivary glands to determine the presence of insulin; none was found. On the other hand they confirmed the presence of

peroxydases and oxydases, so that the secretion of the salivary glands would be able to replace that of the pancreas; this would explain the hypertrophies of the salivary glands observed in diabetics. As a practical result of these observations, in the treatment of diabetes the secretion of the pancreas must be taken into account, and if there is a deficiency of this secretion, adequate substitute preparations should be given.—G. L. Banay.

Hypoglycemia as a cause of mental symptoms. Report of cases. Greenwood, J. Jr., Pennsylvania M. J. **39:** 12. 1935.

This article consists essentially of a discussion of 6 cases in each of which the outstanding symptoms were mental, with an apparent hyperinsulinism as the background. The author believes that there is a somewhat higher percentage of low blood sugar determinations in psychopathic subjects than in patients in the general and medical surgical wards. The significant features are: (1) A psychosis which may take almost any form, often violent, with evidences of sensorium involvement; (2) psychotic manifestations alternating with periods of normal behavior, a more or less transient psychosis, depending upon the severity of the hypoglycemia; (3) a low blood sugar estimate during the psychotic period; (4) relief of symptoms when glucose is given; (5) a high glucose tolerance as indicated by a glucose tolerance curve which is below normal either during the period of fall or at any point in its course; (6) a tendency for the psychotic episodes to occur in the early morning, late at night, or occasionally just before meal time. It is suggested that a further study may cast light on some of the obscure and transient psychoses which disappear on regular hospital feeding. Starvation or irregularity in meals seemed to be a precipitating factor in 3 of the 6 cases reported. The symptoms are so quickly relieved by the oral or intravenous administration of glucose that confinement in a mental hospital could be avoided except in a few of the more severe cases. It is interesting to note that convulsions occurred in only 1 of the 6 cases reported.—I. B.

Reduction of the islets of Langerhans in the pancreas of fish by means of diet, overheating and lack of exercise. Hess, W. N., J. Exper. Zoöl. **70:** 187. 1935.

Recently hatched rainbow trout were divided into groups of 300 each and fed the following diets: (1) beef liver; (2) beef liver plus 10% of lard; (3) beef liver plus 10% lard and 20% cooked corn starch. In diets 4 to 6, inclusive, pig spleen was substituted for liver, and in diets 7 to 9, inclusive, beef heart was substituted. Some groups of fish were fed three times, some six times and others eight times daily. In some troughs water was regulated to flow 1 gallon, and in others 12 gallons per minute. After 120 days the islets in the pancreases of uniform-sized fish (52 mm.) from all groups of fish were counted. It was concluded that fatty degeneration of the pancreas accompanied by reduction of islets, as well as of acinar tissue, could be induced in rainbow trout by using fat, or fat and carbohydrate, in the diet, by overeating, and by lack of exercise. Fat and carbohydrate in the diets were responsible for greatest reduction of islets but overeating was responsible for greater reduction of islets than lack of exercise. As the fatty degeneration of the pancreas increased there appeared to be a corresponding lessened blood supply to this organ.—Author's Abst.

Contribution to the study of diabetes mellitus based on investigation of capillary and venous blood sugars (Contribution à l'étude du diabète sucré basée sur les recherches de la glycemie capillaire et veineuse. 2me Memoire). Landau, A., Mme. T. Hercenberg and I. Beiless, J. de physiol. et de path. gén. **33:** 5445. 1935.

Changes in capillary and venous blood sugars in human diabetics were determined by the technique given in the previous paper of this series. It was felt that differences between capillary and venous sugars (D.C.V.) were important indicators of peripheral tissue, i.e. skeletal muscle, use or storage of carbohydrate. Repeated determinations were made after injection of 50 gm. glucose (30 expts.), insulin (18 expts.) and adrenalin (11 expts.). Sugar injections gave typical diabetic sugar values with excessive hyperglycemia, slow rise and fall and an absence or diminution of the subsequent hypoglycemia found in normal subjects. The D.C.V. was increased in 26 of the 30 cases and was greatest coincidental with the capillary peak in 18 of these. There was no relation of the D.C.V. to the clinical condition nor to the initial sugar level. Insulin-treated diabetics showed no definite correlation between the magnitude of the D.C.V. and the drop in blood sugar. This suggested that insulin did not produce its effect by increasing skeletal muscle utilization of sugar. In hypoglycemia the D.C.V. sometimes was negative, i.e. venous values were higher than capillary by addition of sugar from the muscles. Adrenaline increased capillary sugar and also increased utilization in

7 of 11 cases. The authors conclude that peripheral utilization of sugar, as shown by capillary and venous differences, may be independent of insulin and adrenaline.—R. L. Z.

Observations on a dog maintained for five weeks without adrenals or pancreas. Long, C. N. H., and F. D. W. Lukens, Proc. Soc. Exper. Biol. & Med. **32:** 392. 1934.

One male dog was totally depancreatized and after stabilization on a measured diet and 20 units of insulin the left adrenal was removed 11 days later. The insulin requirement remained unchanged after this. Six weeks later the right adrenal was removed and the animal lived 35 days. After the last operation only 4 units of insulin a day were required with an even higher diet to keep the glycosuria at the previous level. On the third and fourth days of a period of insulin withdrawal the D/N ratios were 1.6 and 1.7 respectively (on meat diet only). Cortical extract (Parke, Davis & Co.) was given in doses of 10 cc. daily after the second adrenal was removed. The animal died suddenly with a subdiaphragmatic abscess and peritonitis but was lively and well until 2 days before death with only moderate weight loss. In this case a strikingly small insulin requirement was demonstrated. The survival period of such an animal without insulin remains to be determined.—Authors' Abst.

Effect of adrenalectomy and hypophysectomy upon experimental diabetes in the cat. Long, C. N. H., and F. D. W. Lukens, Proc. Soc. Exper. Biol. & Med. **32:** 743. 1934.

In this series of experiments the survival and the urinary glucose, nitrogen and acetone bodies of 3 normal, 4 depancreatized, 4 hypophysectomized-depancreatized and 5 adrenalectomized-depancreatized cats have been quantitatively compared. Variable doses of cortical extract were given to the adrenalectomized group of animals. Insulin was not given to any animals. The usual high excretion of glucose and nitrogen was much reduced in the doubly operated animals, while ketonuria was almost entirely absent. The absence of ketosis would seem to be the cause of the survival of these animals. Such observations have not previously been made in adrenalectomized-depancreatized animals. They afford a new mechanism by which the Houssay type of modified pancreatic diabetes may be produced.—Authors' Abst.

Complications of diabetes mellitus. McMullen, C. J., Illinois M. J. **68:** 327. 1935.

The author enumerates the causes of death in a series of 130 fatal cases of diabetes mellitus out of 600 cases seen in the Cook County Hospital during the year 1933. The chief complications of diabetes mellitus are coma, infections including pulmonary tuberculosis, gangrene, arteriosclerosis, and heart disease. The death incidence according to age was as follows: 1st decade—1; 2nd decade—5; 3rd decade—4; 4th decade—11; 5th decade—25; 6th decade—31; 7th decade—34; 8th decade—18; 9th decade—1. Of the 130 fatalities, 36 were due to coma. Of these, 27 lived 1 day, 5 lived 2 days, 3 lived 3 days, and 1 over 3 days. If the patient is in deep coma, he usually dies within 24 hours. If he lives 48 to 72 hours after entrance into coma, he will probably recover under adequate treatment. Of 23 diabetic autopsies, active pulmonary tuberculosis was present in 2 cases. In 20 cases of diabetes in children, 6 showed evidence of tuberculosis. In order to prolong the life of these patients the clinician should be thoroughly acquainted with the complications of diabetes.—I. B.

The response of the canine and human pancreas to secretin. Voegtlin, W. L., H. Greengard and A. C. Ivy, Am. J. Physiol. **110:** 198. 1934.

In the dog and in man there is no consistent increase in the concentration of pancreatic enzymes found in the duodenal drainage after the intravenous injection of secretin. In tests made on 22 human subjects a simultaneous rise in the concentration of amylase, lapase and trypsin in the duodenal contents after secretin injection was noted in only 1 instance. In 6 instances, the concentrations of these enzymes were simultaneously decreased. In the remaining 15 tests the concentrations of these enzymes were increased or decreased in an apparently random fashion. The total output of all the enzymes (trypsin, amylase, lipase) is almost invariably increased during the first 15 minute period after secretin injection in both the dog and in man. With the doses of secretin used, the fluid output should be nearly tripled during the 15 minute post-injection period. The results of secretin stimulation must be interpreted from an increased total production of enzymes and fluid. This fact is unfortunate in that it requires for absolute quantitative conditions, at least, a quantitative drainage of the duodenum, a condition probably not attained by the present methods of duodenal intubation. The average, maximum and minimum values for the production of enzymes and fluid before and after

secretin injection are given in the text. Repetition of the test on two subjects resulted in stimulation of the pancreas in both instances but did not result in strictly constant results. In other words, the response of the same individual to secretin varies from time to time. A tentative procedure for testing the pancreatic response to secretin together with our interpretation of the results obtained has been given. In view of the severe reaction that occurred in two subjects on receiving a second injection more than one year after the first injection, it is believed that this preparation of secretin should be injected only once. While purposefully no pathological cases were included in this work, it is believed that the test has pointed out two cases of pancreatic deficiency in subjects that were presumed to be normal. However, it is believed that it has yet to be established that any test based on duodenal drainage is of more practical value than stool analysis. This statement is made to encourage, rather than to discourage, further investigation in this field.—Authors' Summary.

Hyperparathyroidism due to diffuse hyperplasia of all parathyroid glands rather than adenoma of one; clinical studies on three such cases. Albright, F., Esther Bloomberg, B. Castleman and E. D. Churchill, Arch. Int. Med. **54:** 315. 1935.

Three patients with clinical hyperparathyroidism were found at operation to have multiple parathyroid enlargements, considered to represent hyperplasia of all parathyroid tissue. The histologic structure of the glands in these patients differed markedly from that in 16 cases from the clinic in which only solitary parathyroid enlargements were present. The distinctive features were the uniformity of structure, the enormous size of the cells, the extreme clearness of the cytoplasm and the tendency to glandular formation. A distinction is made between parathyroid hyperplasia with hyperparathyroidism and compensatory hyperplasia of the parathyroid glands. An analogy between the former and exophthalmic goiter is made. Hyperplasia of the parathyroid glands with hyperparathyroidism is considered a disease entity. The surgical treatment of parathyroid hyperplasia with hyperparathyroidism brings up some interesting questions. Thus removal of two enlarged glands in one of these patients was without effect on the serum calcium and inorganic phosphorus levels. Even with removal of sufficient tissue to alter the blood chemistry satisfactorily, the condition is apt to recur. It is still undecided whether this condition can be handled by surgical means. A review of 101 cases of hyperparathyroidism revealed 17 cases, including these 3, in which there were multiple parathyroid enlargements. Several are undoubtedly similar to the ones here reported. The records of the cases of multiple parathyroid tumors have abundant circumstantial evidence in them to suggest that the pituitary gland is incriminated. The cause of the hyperplasia is probably a parathyroid-stimulating factor, possibly the one of the anterior lobe of the pituitary described by Hertz and Kranes.—Authors' Summary.

Influence of thyro-parathyroidectomy and of parathyroid hormone upon state of calcium in serum of the cat. McLean, F. C., B. O. Barnes and A. B. Hastings, Proc. Soc. Exper. Biol. & Med. **32:** 253. 1934.

Hypo-calcemia following thyro-parathyroidectomy results in a diminished concentration of Ca ions, and hypercalcemia following administration of the parathyroid hormone results in a correspondingly increased concentration of Ca ions. No evidence of any effect of removal of the parathyroid glands, or of administration of the parathyroid hormone, upon the qualitative state of calcium in the serum was found, the changes observed being simply the quantitative changes predicted by the mass-law equation. Symptoms of tetany usually appeared at a Ca ion concentration of approximately 0.6 mM per kg. H_2O, increased in severity at still lower levels, and disappeared as the Ca ions concentration rose again above the 0.6 mM level.—M. O. L.

Hyperparathyroidism and angiospastic syndromes (Hyperparathyroïdie et syndromes angiospastiques). Bastai, P., and G. C. Dogliotti, Presse méd. **47:** 1766. 1934.

Clinical observations were made on the results of parathyroid operation in 4 cases of endarteritis and diabetic endarteritis. After successful parathyroidectomy the authors observed the following facts: (1) Calcemia regressed to almost normal level; (2) pains, even those of long duration, ceased immediately; (3) subjective and objective increase of the local temperature of the affected parts and increase of the sudoral secretion; (4) delimitation and surprisingly rapid cure of the wounds which had previously resisted treatment for many years; (5) in cases of hypertension, lowering of the arterial pressure. The authors try to correlate these findings with the function of the parathyroid.—G. L. Banay.

Studies on physiology of the parathyroid glands. Ellsworth, R., and J. E. Howard. Bull Johns Hopkins Hosp. **55:** 296. 1934.

Six individuals in whom the kidneys gave evidence of functioning normally were kept fasting, at rest, and on a given fluid intake. Urine was collected hourly for 3 hours before and 3 hours after the intravenous injection of 40 units of parathormone. Following the extract there was usually, but not always, a diuresis. There was no alteration in urea clearance. There was a tendency for the pH to shift toward the alkaline side. The chloride excretion usually rose very slightly in the first hour after the injection and then decreased quite markedly in the second and third hours. Most striking was the out-pouring of phosphate in the urine. This was very conspicuous during all 3 hours after the parathormone. The phosphate clearance in cc. of plasma cleared per hour increased from two to five-fold after the injection. The phosphorous diuresis was accompanied by a relatively slight fall in serum inorganic phosphate. A definite effect of intravenous parathyroid extract upon the normal kidney is indicated, most conspicuously apparent in the excretion of phosphate. It is similar to the effect of intramuscular injection in hypo-parathyroid patients previously reported.—J. E. Howard.

The influence of the parathyroid on the metabolism of creatine and phosphoric acid. Imrie, C. G., and C. N. Jenkinson. J. Physiol. **79:** 218. 1933.

In thyroparathyroidectomized cats the creatine phosphate content of the limb muscles was measured before and after stimulation. The concentration of creatine phosphate was noted without the administration of parathormone, with the administration of parathormone, and with creatine administered along with parathormone in feeding experiments. It was found that the creatine phosphate concentration was lower and the rate of resynthesis slower following muscle stimulation in these animals than in normal ones. The resynthesis after the administration of parathormone took place at a rate similar to that observed in normal animals. When creatine was administered by mouth along with parathormone the initial values were higher than the normal average and the rate of resynthesis after muscle stimulation followed the normal curve.—J. S. Gottlieb.

Clinical treatment of certain bone diseases caused by hyperfunction of the parathyroid glands. Wagoner, G., Pennsylvania M. J. **37:** 983. 1934.

The systemic damage resulting from hyperfunction of the parathyroid glands can be checked, alleviated, and in part repaired by proper therapeutic measures. These measures comprise: (1) roentgen irradiation of the parathyroids; (2) a diet containing adequate amounts of calcium and phosphorus in proper ratio, supplemented by vitamin D; and (3) the surgical removal of one or more parathyroid glands. These agencies may be used singly or in combination. It is imperative that before instituting remedial measures, accurate and thorough chemical studies of the patient's calcium and phosphorus metabolisms should be made. These determinations should be repeated at frequent intervals throughout treatment. —I. B.

Obesity treatment by diet, thyroid and dinitrophenol; result on 106 outpatients. Bayer, L. M., and H. Gray, Am. J. M. Sc. **189:** 86, 1935.

The methods used in treating 106 unselected obese patients in the outpatient clinic were analyzed, with reference to the effects of diet alone (100 cases), thyroid (51) and dinitrophenol (23). The weight losses shown here are not so great with any of these methods as could have been achieved if patients had restricted their diets more conscientiously. They do, however, represent the sort of results which may be expected in outpatient care of unselected patients, and, therefore, permit the following tentative practical conclusions: (1) Diet alone will take off an average of 15 pounds in $3\frac{1}{2}$ months and, therefore, should be the first therapeutic procedure in the treatment of obesity. When diet fails to get results, either thyroid or dinitrophenol appears to be an effective adjunct. As an adjunct to diet, dinitrophenol is at least as effective as thyroid and, in patients with normal basal metabolism, possibly superior. Thyroid given immediately with the beginning of diet caused no better loss than when it was given only after loss on diet had stopped. Dietary control was necessary to achieving results with dinitrophenol, as it was to getting results with thyroid. Furthermore, reasoning from the thyroid results by analogy, it would seem that it is worth while removing as many pounds as possible on diet alone before treating with dinitrophenol—this on the assumption that with any drug which is not compensating a physiological deficiency, the less given, the better for the ultimate health of the patient. It should be emphasized that neither

thyroid nor dinitrophenol should be used indiscriminately by patients unguided or under the direction of physicians inexperienced with their use.—Authors' Summary.

Contribution to the question of the antithyreoidin dosage. Breitmann, M. J., Acta endocrinol. ucrainica pp. 110-111. 1934.

In severe cases of Graves' disease antithyreoidin is of no value. In mild cases of hyperthyreosis it is very useful: it relieves the insomnia, tremor and tachycardia. Its combination with insulin is beneficial. Small doses (10-15 drops) are of no use whatever. Antithyreoidin should be given once a day, one hour before sleep, 3-5 grams in soda water for 3-5 days, periodically if necessary.—Author's Abst.

Hypothyroidism and cretinism in childhood. IV. Post mortem report on two cretins. Bronstein, I. P., and G. Milles. Am. J. Dis. Child. **49:** 1565. 1935.

The autopsy findings in two cases of cretinism in 4th and 8th month old children disclosed no trace of thyroid tissue on either macroscopic or microscopic examination. In 1922, Gordon found only four authentic cases of athyreosis in the American literature.—M. B. G.

Sporadic cretinism in one of twins. Report of cases with roentgen demonstration of osseous changes that occurred in utero. Dorff, G. B., Am. J. Dis. Child. **48:** 1316. 1934.

Two cases are presented in which sporadic cretinism occurred in two sets of differently sexed twins, one twin in each set being involved, the other twin being normal in all respects. Definite X-ray evidence is given that the retarded osseous development in a twin with congenital sporadic cretinism occurred in utero. The author believes that: a) there is an independent, low grade fetal thyroid activity which is not directly influenced by the maternal thyroid hormone; b) there is probably no transmission of the thyroid hormone through the placenta from either the fetus or mother.—M. B. G.

Therapeutic effect of total ablation of normal thyroid on congestive heart failure and angina pectoris; relationship between serum cholesterol values, basal metabolic rate and clinical aspects of hypothyroidism. Gilligan, D. R., M. C. Volk, D. Davis and H. L. Blumgart, Arch. Int. Med. **54:** 746. 1934.

Simultaneous measurements of the serum cholesterol concentration and basal metabolic rate have been made in a group of patients before and at intervals during the development of the hypothyroid state after total ablation of the normal thyroid gland. Following total thyroidectomy, the serum cholesterol value increases as the basal metabolic rate decreases and as the clinical evidences of hypothyroidism become manifest. The highest serum cholesterol concentration obtained in any patient was 543 mg. per hundred cubic centimeters and the lowest basal metabolic rate was minus 47%. Appreciable changes in the basal metabolic rate and serum cholesterol concentration were generally evident by the end of the first postoperative week. Striking changes in the values were observed by the end of the first postoperative month. During the following few months the basal metabolic rate showed only minor, if any, decreases, whereas the serum cholesterol concentration gradually increased and the clinical course of hypothyroidism gradually progressed. When the untoward symptoms of myxedema became manifest, the serum cholesterol concentration was usually increased to 300 mg. per hundred cubic centimeters or above, and the basal metabolic rate was usually decreased to minus 30% or below. Small doses of thyroid medication, administered to control the untoward symptoms of myxedema, caused slight increases in the basal metabolic rate and significant decreases in the serum cholesterol concentration. In a few instances a high serum cholesterol value confirmed the clinical finding of untoward symptoms of myxedema when the basal metabolic rate was not markedly low. In other instances, when duplicate measurements of the basal metabolic rate which agreed within 5% could not be obtained, the serum cholesterol concentration was a valuable adjunct to clinical impressions in estimating the degree and course of the hypothyroid state.—Authors' Summary.

Basic amino acids yielded by thyroglobulin. White, A., and W. G. Gordon, Proc. Soc. Exper. Biol. & Med. **32:** 354. 1934.

Analysis of the basic amino acids of thyroglobulin by direct isolation methods indicates that this protein yields 0.62% of histidine, 8.22% of arginine and 1.93% of lysine.—Authors' Summary.

The use of iodine in recurrent exophthalmic goiter. Haines, S. F., West. J. Surg. **42:** 449. 1934. Abst., J. Nerv. & Ment. Dis. **81:** 73.

During the years 1928 to 1932 inclusive, 488 patients with recurrent or persistent exophthalmic goiter were examined in The Mayo Clinic. In 123, or 25.2% of these cases, medical treatment with compound solution of iodine brought about satisfactory control of the disease. The average basal metabolic rate before the original thyroidectomy was +48.66%, before treatment of the recurrence +20.98%, and after treatment of the recurrence —1.56%. Seventy-three patients each received iodine for more than a year, and 23 patients took iodine continuously, or nearly so, for between 1 and 5 years. Several patients took iodine for the greater part of the time for several years and, judging from their histories, had recurrences of exophthalmic goiter whenever they stopped using it for any considerable time. The effect of iodine was obtained repeatedly in many cases. No difficulties were encountered in this group of cases as a result of long continued use of iodine, but it must be noted that in each case in the group the disease was well controlled before the decision was made to continue with medical treatment alone. The thyroid glands were slightly enlarged to palpation in only 45 cases. In no case in this group was there great enlargement of the thyroid gland. The amount of gland tissue palpable at the time of recurrence is probably of more value than any other single fact in helping to predict the possibility of control with iodine.

Blood cholesterol and creatine excretion in the urine as aids to diagnosis and treatment of hypothyroidism. Hess, J. H., Ann. Int. Med. **8:** 607. 1934.

The blood cholesterol was determined in 25 children ranging from 2 months to 11 years of age. Three of the group were normal, the rest were presumably free from thyroid deficiency but suffered from various disorders from severe malnutrition to obesity. The mean cholesterol value was 190 mg. per 100 cc. of blood, with a range from 129 to 217 mg. %. In 8 boys and 4 girls the blood cholesterol ranged from 277 to 782. These values were reduced by thyroid therapy. In two children suffering from thyroid deficiency the normal prepubertal creatinuria was reduced or completely absent. Thyroid feeding caused a resumption of the creatinuria which occurred before any definite change is noted in B.M.R. or blood cholesterol. —J. M. L.

The interaction of food and sanitary condition in the causation and prevention of thyroid disease. McCarrison, R., Indian J. M. Research, **20:** 957. 1933.

Seven groups of 36 young rats each were reared under the following conditions:— Group I: good sanitation and superior diet. Group II: good sanitation and inferior diet. Group III: bad sanitation and superior diet. Group IV: bad sanitation and inferior diet. Group V: good sanitation, superior diet and unsterile filtrate of excreta and decaying food. Group VI: good sanitation, superior diet and sterile filtrate. Group VII: good sanitation, superior diet and sterile filtrate plus iodine. The well-constituted diet, described as "superior," had definite anti-hyperplastic or anti-goitrogenic, qualities; this diet afforded albino rats, subjected to the above conditions of insanitation, a high degree of protection against hyperplastic goitre. In the compost of excreta and decaying food accumulating under these conditions of insanitation there existed a water-soluble and water-borne "noxa" which, when administered per os in daily doses of 10 cc. of the watery solution, caused retrograde changes, of an apparently adenomatous nature, in the thyroid gland of albino rats. These retrograde changes were not prevented by the well-constituted ("superior") diet; nor by the administration of iodine simultaneously with the watery solution of the noxa. The goitre-noxa associated with insanitary condition may be of dual nature: comprised of a hyperplasia-producing factor and an adenoma-producing factor. The former is counteracted by a well-constituted diet and by iodine; the latter (in the doses in which it was administered) is not. Or, the noxa may be a single substance whose action on the thyroid differs with its dosage: its ingestion in larger amounts over long periods causing retrograde changes in the organ even in the presence of a well-constituted diet and of iodine; its ingestion in smaller amounts causing hyperplastic goitre but only in the presence of an ill-constituted diet. The interaction of food and sanitary condition on the causation or prevention of thyroid disease is again made evident by the results here recorded; and the limitations of a perfectly constituted diet and of iodine in counteracting this goitre-noxa are indicated.—R. G. H.